BioEnergy Science Center (BESC)

Director’s Presentation to BERAC

Paul Gilna

February 27, 2015

The BioEnergy Science Center

A multi-institutional, DOE-funded center performing basic and applied science dedicated to improving yields of biofuels from cellulosic biomass

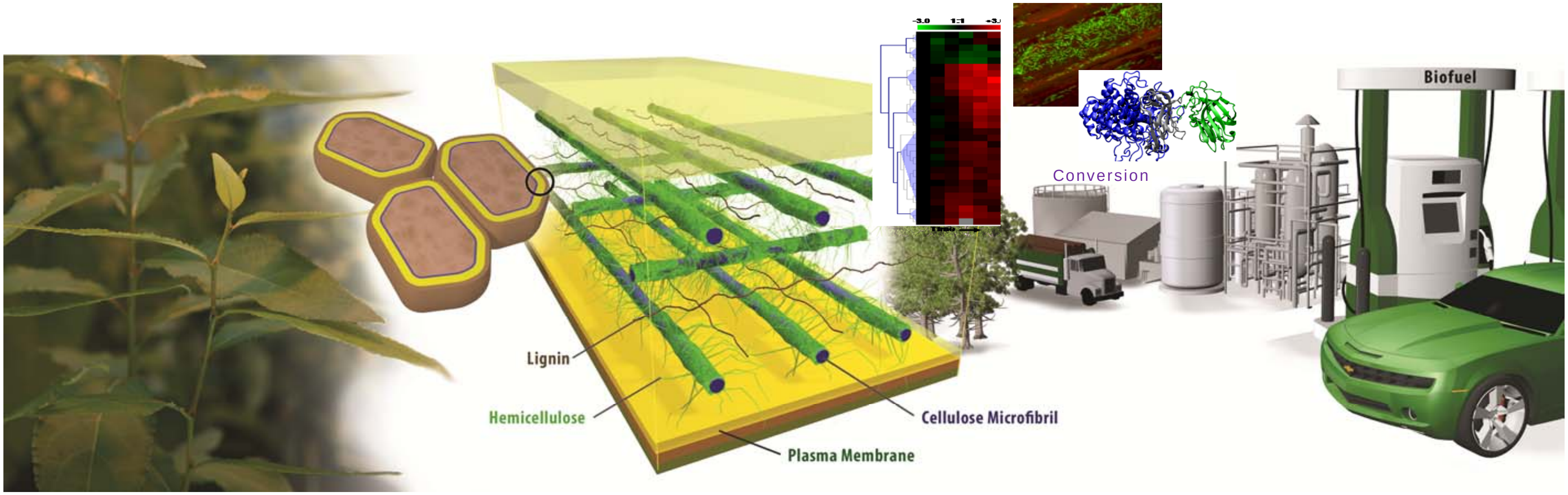
300+ People in 18 Institutions

- Oak Ridge National Laboratory
- National Renewable Energy Laboratory
- Samuel Roberts Noble Foundation
- ArborGen, Inc.
- Ceres, Incorporated
- Mascoma Corporation
- DuPont
- GreenWood Resources
- University of North Texas

- University of Georgia
- University of Tennessee
- Cornell University
- Dartmouth College
- West Virginia University
- Georgia Institute of Technology
- University of California--Riverside
- North Carolina State University
- University of California—Los Angeles

2012-2014

BESC mission statement



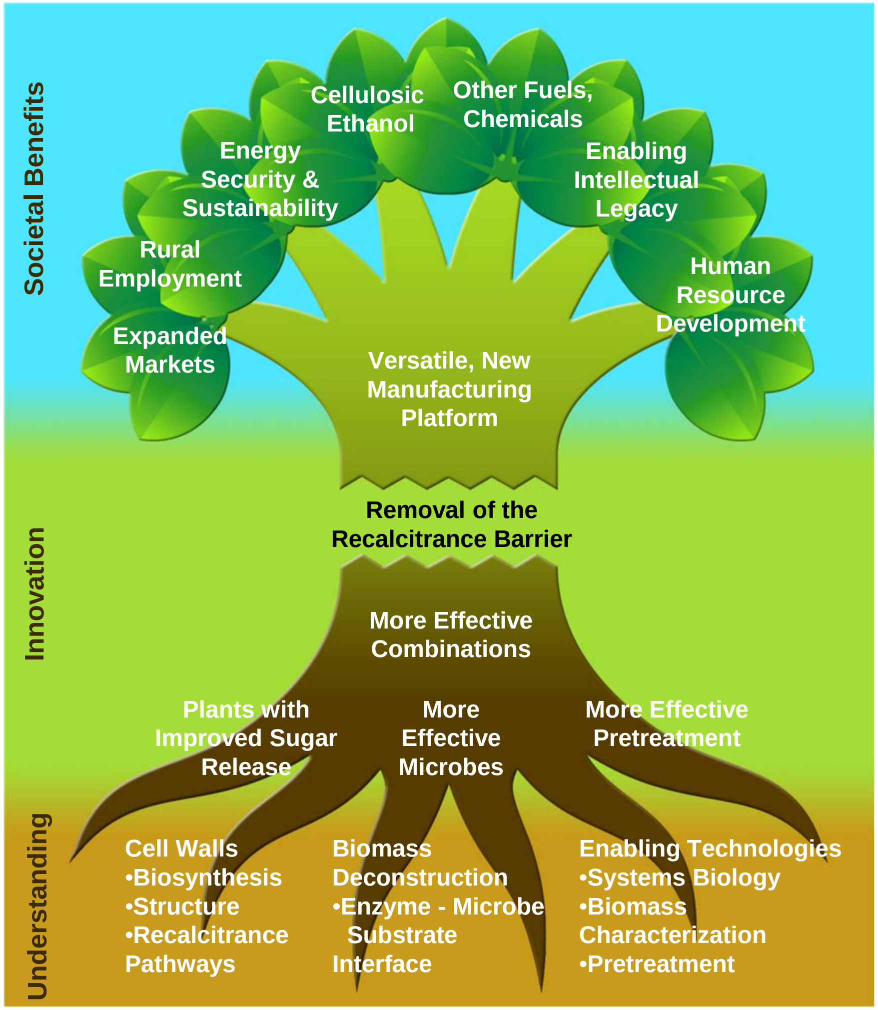
To enable the emergence of a sustainable cellulosic biofuels industry by leading advances in science and science-based innovation resulting in removal of recalcitrance as an economic barrier to cost-effective production of biofuels.

Access to the sugars in lignocellulosic biomass is the current critical barrier

- Overcoming this barrier will cut processing costs significantly and be used in most conversion processes.

- This requires an integrated, multi-disciplinary approach.

- ***BESC believes biotechnology-intensive solutions offer greatest potential.***



Societal Benefits

Cellulosic Ethanol

Other Fuels, Chemicals

Energy Security & Sustainability

Enabling Intellectual Legacy

Rural Employment

Human Resource Development

Expanded Markets

Versatile, New Manufacturing Platform

Innovation

Removal of the Recalcitrance Barrier

More Effective Combinations

Plants with Improved Sugar Release

More Effective Microbes

More Effective Pretreatment

Understanding

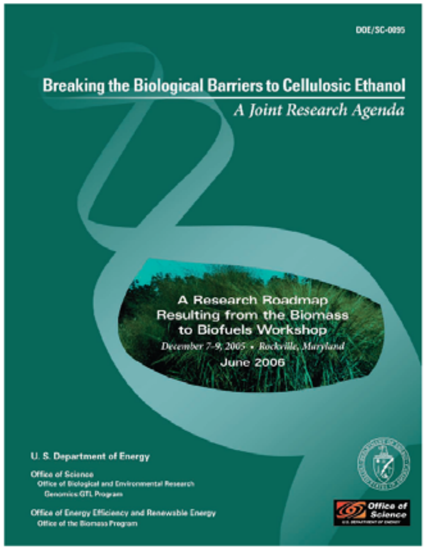
Cell Walls
•Biosynthesis
•Structure
•Recalcitrance Pathways

Biomass Deconstruction
•Enzyme - Microbe Substrate Interface

Enabling Technologies
•Systems Biology
•Biomass Characterization
•Pretreatment

Scientific challenges as of 2006

- With a deeper understanding of:
    - The resistance of lignocellulosic biomass to deconstruction
    - The genetic controls of plant composition and ultrastructure
    - Bioenergy crop domestication and sustainability
    - The structure and function of cellulases and other plant cell wall depolymerizing enzymes
    - The microbial cell's mechanisms for toxicity response
- We could envision:
    - Dedicated bioenergy crops
    - Consolidated bioprocessing – cellulase production and ethanol fermentation combined
    - Beyond "ethanol" to advanced biofuels
    - Improved pretreatments



DOE/SC-0095

Breaking the Biological Barriers to Cellulosic Ethanol

A Joint Research Agenda

A Research Roadmap Resulting from the Biomass to Biofuels Workshop

December 7–9, 2005 • Rockville, Maryland

June 2006

U. S. Department of Energy

Office of Science
Office of Biological and Environmental Research
Genomics GTL Program

Office of Energy Efficiency and Renewable Energy
Office of the Biomass Program

DOE Biomass to Biofuels Roadmap (7/2006)

http://doegenomestolife.org/biofuels/b2bworkshop.shtml

Cellulosic industry is emerging

Syngenta and Cellulosic
Ethanol Technologies team
up for
in Iowa
July 14, 201

In Minnes
successfu
ethanol—
Processo
one millio
in 2015. E

Read M

M&G Chemicals, Anhui
Guozhen ink pact to build
world's
biofuel
July 29, 201

970,000-1
30% per g
Chemical
PROESA
970,000-1
cellulosic

Read M

Walking The Talk: A
fast-revving DuPont nears
completion of its cellulosic
bioref
Septembe

Sustain
what do
critical,
2002, D
Schmidt
Watts (t

Read

Editor's Sketchbook: Beta
Renewables cellulosic
ethanol project
Crescentino, It
September 25, 2014 | Jim

The first thing you no
plant in Crescentino i
away" era of cellulosi
small shacks that hel
project, up close and

Read More

GranBio starts cellulosic
ethanol production at 21
million gallon plant
Alagoas, Brazil
September 24, 2014 | Jim Lane

World's largest-known cell
in Brazil in record time. In
the first commercial-scale
Southern Hemisphere. Th
Campos, Alagoas, has an
of ethanol per […]

Read More

Beta Renewables,
Biochemtex ink deal for
commercial-scale c
biofuels project in S
October 6, 2014 | Jim Lane

Cellulosic leaders sign with
2nd Generation Ethanol pla
Biochemtex and Beta Rene
Energochemica SE for the c
ton per year) cellulosic etha
constructed in Strazske, Slo

Read More

World's largest cellulosic
biofuels plant opens: Beta
Renewables, in pictures
October 10, 2013 | Jim Lane

Beta Renewables plant in Crescentino,
Italy to produce 75 million liters of
cellulosic ethanol per year from agricultural waste. What does a
commercial-scale plant look like? In Italy, Beta Renewables and
Novozymes marked the official opening in Northern Italy of the world's
largest cellulosic biofuels facility. Situated in fields outside the city of
Crescentino, it […]

Read More

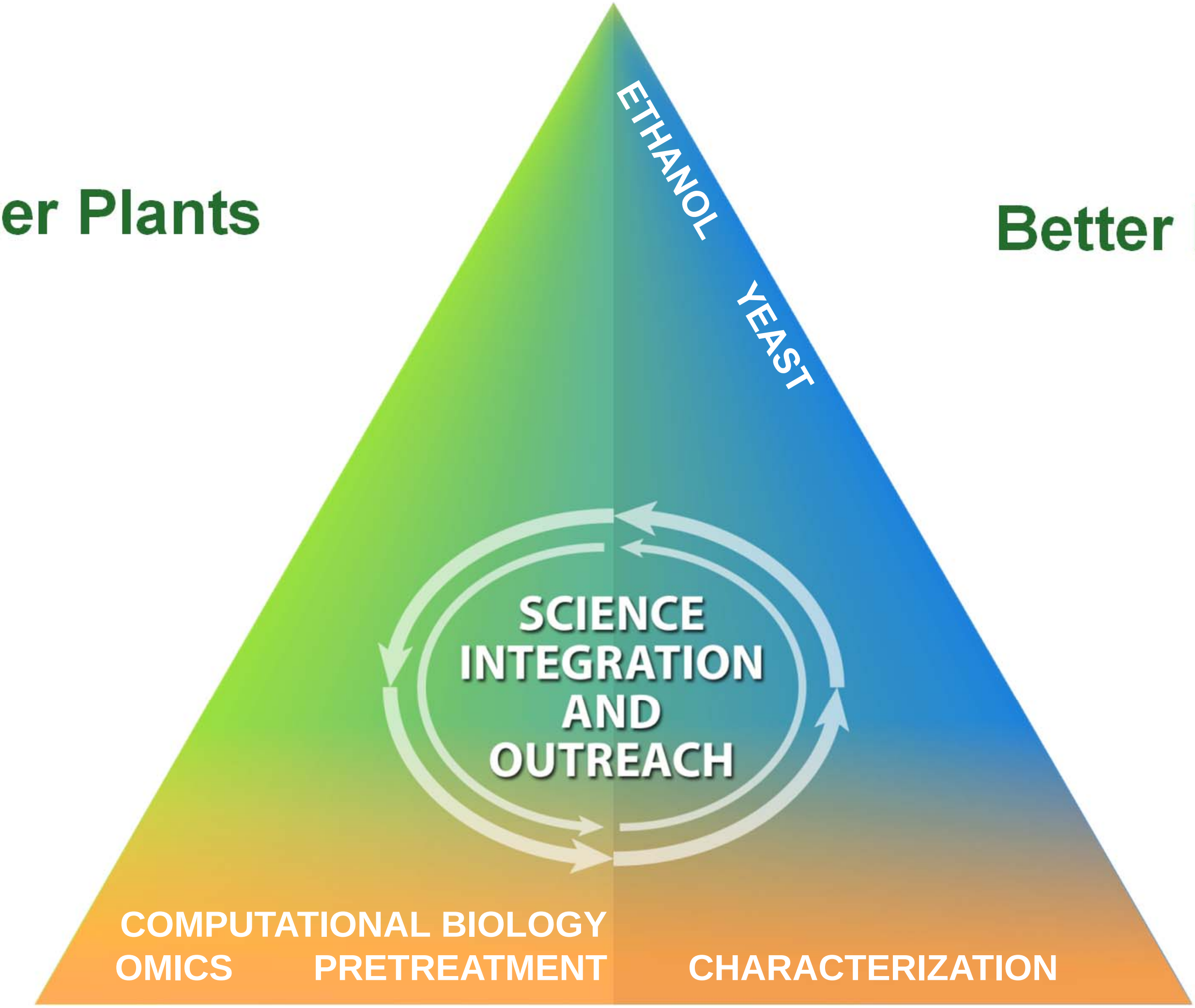
Key strategic goals for BESC through Year 10

- Develop a fundamental understanding of the molecular basis of recalcitrance and cell wall formation.
- Identify and characterize a high-performing set of *Populus* and switchgrass TOP reduced recalcitrance lines.
- Understand microbial cellulose utilization at all levels, including enzyme components, enzyme systems, pure cultures, and mixed cultures.
- Achieve proof of concept for CBP with *C. thermocellum,* yeast, and *Caldicellulosiruptor spp.*
- Develop and apply chemical, immunological, physical, and imaging methods to characterize biomass; use the resulting data to build models to predict the relationships between biomass structure and recalcitrance.

Organized around our strategic goals

Better Plants

Better Microbes



ETHANOL

YEAST

SCIENCE INTEGRATION AND OUTREACH

COMPUTATIONAL BIOLOGY

OMICS PRETREATMENT

CHARACTERIZATION

ENABLING TECHNOLOGIES

Better Tools and Combinations

Feedstocks: then and now

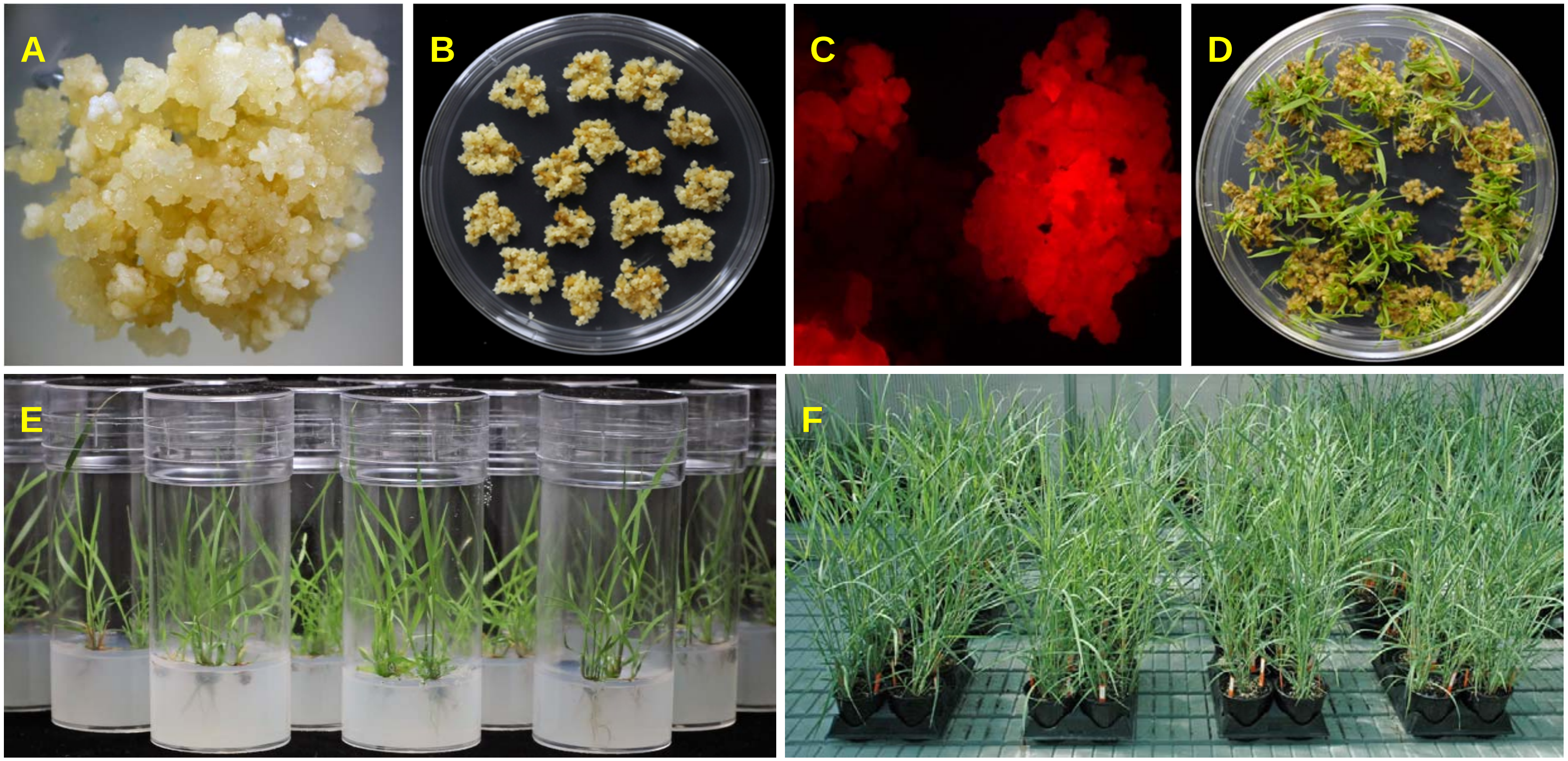
2007

- Lignin and cellulose accessibility believed to be the primary roots of recalcitrance
- Low transformation efficiencies for switchgrass
- Range of natural variation and genetic control of recalcitrance within a species not established
- Viability of low lignin transgenic perennials not confirmed in field

2015

- Proved core concept that multiple genes – many outside of the lignin pathway, control plant cell wall recalcitrance
- Used BESC transformation pipeline to target ~900 candidate recalcitrance gene constructs to date
- Verified multiple reduced recalcitrance lines in *Populus* and switchgrass and ~20 TOP Lines selected for deeper study
- Data now returning from field trials of initial transgenic lines

Early results: high-throughput *Agrobacterium*-mediated switchgrass transformation

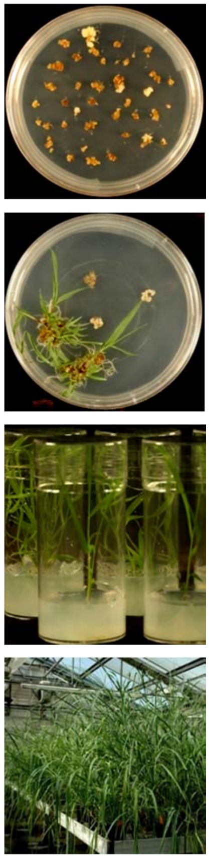


- A transformation efficiency of more than 90% has been achieved, vastly improving prior 5% efficiency.
- The system has been effectively used for producing large numbers of transgenic switchgrass plants.

Early results: COMT modification improves biofuel yield

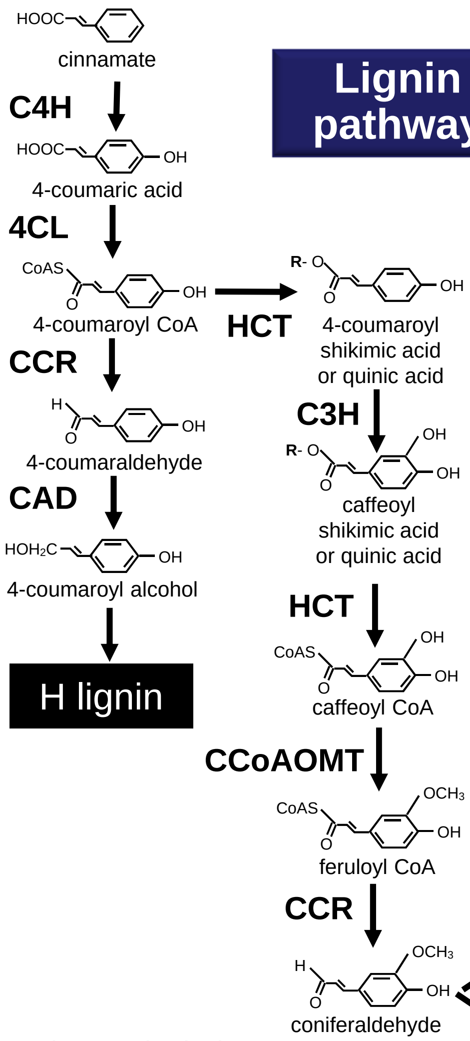
Phenylalanine → **PAL**

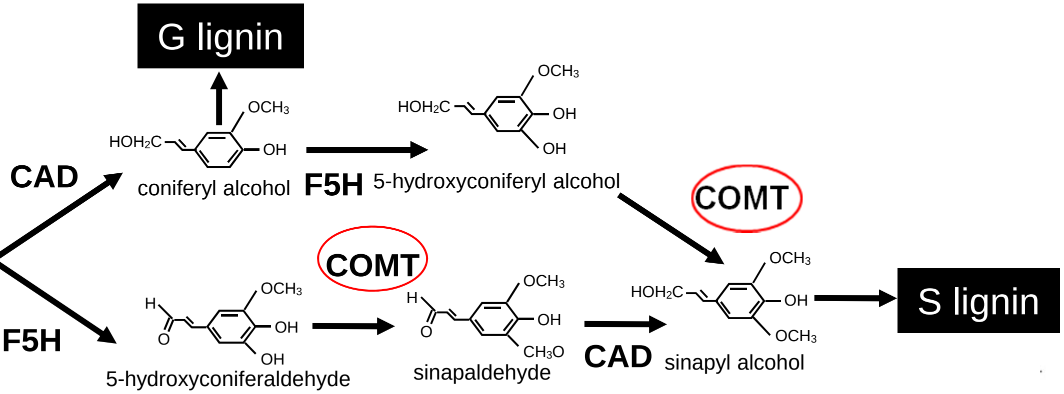
Agrobacterium-mediated transformation of switchgrass

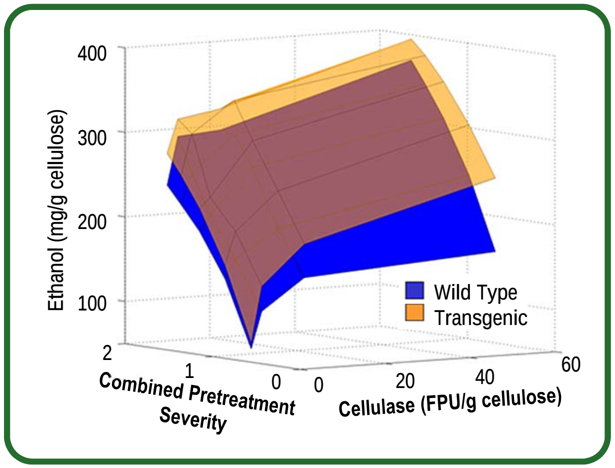


The Samuel Roberts
NOBLE
Foundation

Lignin pathway







Impact of enzyme levels and pretreatment conditions on biofuel production

Noble Foundation transgenic switchgrass

Fu and Wang (Noble),
Mielenz (ORNL),
support from USDA/DOE
Fu *et al*. *PNAS* 2011

Field study of COMT transgenic switchgrass

Significance

This is the first reported field study evaluating biofuel potential of transgenic switchgrass with reduced cell wall recalcitrance and shows that conversion phenotypes can be maintained in the field.

Outcome

- Lignin-modified transgenic switchgrass had similar gains in sugar release (up to 34% higher) and biofuel production (up to 28% higher) as observed in the greenhouse.
- Results extrapolate to 50% more liters of ethanol per hectare compared to wild-type.

BESC Synergy

- Culmination of several years and many hands;
- Prior BESC work: creation of the transgenic switchgrass (Noble), greenhouse experiments (Noble), composition analyses (Noble, NREL);
- This work: APHIS field permits and cultivation (UTK), compositional analyses (NREL, ORNL), saccharification tests (NREL), SHF fermentation tests (ORNL, UTK).

Plant
Biotechnology
Journal

Volume 12 | Issue 7
September 2014

www.plantbiotechjournal.com | ISSN 1467-7644

WILEY
Blackwell

Plant Biotechnology Journal
September 2014 Cover

Baxter, H.L., *et al.*, *Plant Biotech J* 12 (7), 2014.

Genome-wide adaptive variation in *Populus* revealed by population genomic analysis

Significance

- Study is the first to explore the genomic legacy of selection across an entire tree genome and highlight both the wide range of selection pressures as well as the climatic influence on phenological systems and growth.
- Provides new insights into mechanisms of adaptive evolution, and provides potential targets for genetic improvement for biofuels production as well as the management of natural populations in the face of environmental change.

Outcome

- 17.9 million single nucleotide polymorphisms (mostly rare) were identified representing naturally segregating variants found in wild populations which can be utilized to track key traits.
- 379 genomic regions showed evidence of recent positive and/or divergent selection.
- These selected regions had stronger gene-trait association signals at all three test sites than expected, indicating that these outliers were driven by adaptive selection and represent natural, functional variation.

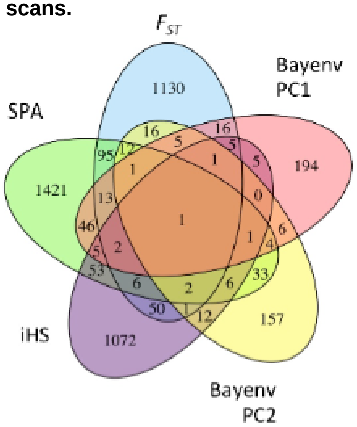
BESC Synergy

- This work builds on several years of groundwork:
  - ➢ establishing the common gardens
  - ➢ JGI sequencing the thousand individuals
  - ➢ multiyear phenotyping team(s)
- This work represents multiple skill sets: field and population design (WVU, ORNL), field maintenance (GreenWood and Mt. Jefferson Farms), plant genome sequencing (JGI, ORNL), and GWAS bioinformatic analytics (ORNL, WVU, UTK)

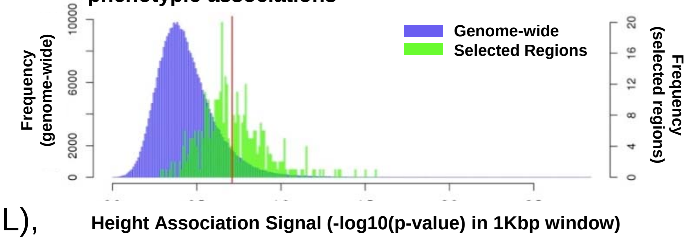
Locations of plantations (yellow diamonds) and tree collections. Colors indicate genetically differentiated groups.

Spring Bud Flush Phenotypes

Venn diagram showing the unique and overlapping regions identified by the five genome-wide selection scans. 379 regions were identified by >2 scans.



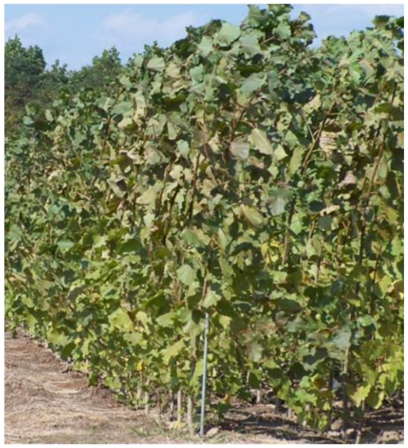
Correspondence between selection scans and phenotypic associations



Evans, *et al*., *Nature Genetics, 46*, 1089-1096, 2014.

Field testing of improved feedstocks

ARBORGEN



40+ *Populus* constructs in stool beds (South Carolina)

OAK RIDGE National Laboratory

1000+ *Populus* genotypes in 4 common gardens (Pacific Northwest)

ceres

Field assessment of genetically improved switchgrass (Texas)

THE UNIVERSITY of TENNESSEE UT

Field assessment of genetically improved switchgrass (Tennessee)

Reduced recalcitrance TOP Lines already selected for deeper study

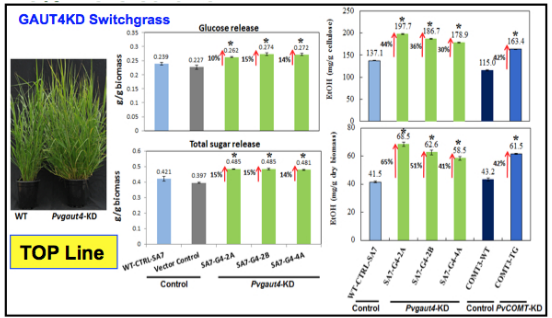
Year 6

- *Pv*GAUT4-KD (pectin synthesis)
- *Pv*FPGS1-KD (carbon metabolism)
- *Pv*CCR1-KD (lignin synthesis)
- *Pv*MYB4-OE (transcription factor, lignin)
- *Pv*COMT-KD (lignin synthesis)
- *Pv*microRNA156-OE (growth and development)

Year 7

- PvSGAMP15_14 (natural variant)
- PvSGAMP15_05 (natural variant)
- PvEFR/SHINE TF-OE (transcription factor)
- PvGAUT1-KD (pectin synthesis)

+18 Populus lines added



Switchgrass control and *FPGS*-KD lines at R1 stage.

Reduced recalcitrance TOP Lines already selected for deeper study

Year 6

- PvGAUT4-KD (pectin synthesis)

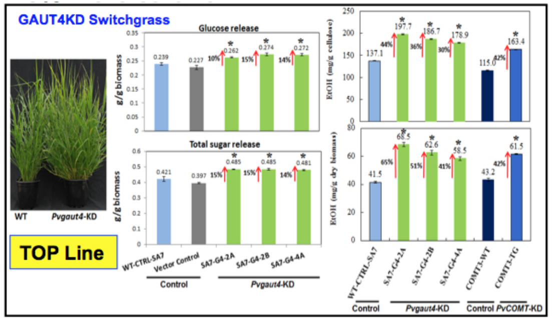
BESC Synergy

- Each of the TOP Lines combine insights from multiple institutions and simple to complex analytics.
- Common shared protocols and metrics allow stronger comparisons.
- Shared improvements in genetics, transformation and field tests accelerate efforts and avoid errors.

gnin)

Ye

- PvEFR/SHINE TF-OE (transcription factor)
- PvGAUT1-KD (pectin synthesis)



Switchgrass control and *FPGS*-KD lines at R1 stage.

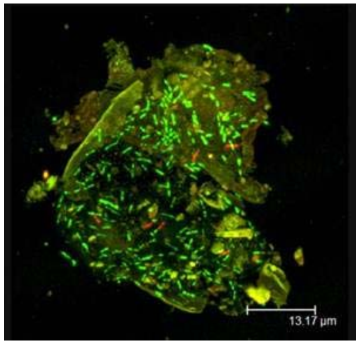
CBP microbes: then and now

2007

- Few cellulases expressed in yeast
- Functional genetic systems for cellulolytic thermophiles not available
- Modes of ethanol inhibition or tolerance in *C. thermocellum* not understood
- *Caldicellulosiruptor* not well characterized
- Few models and mechanisms for multifunctional cellulolytic enzymes available

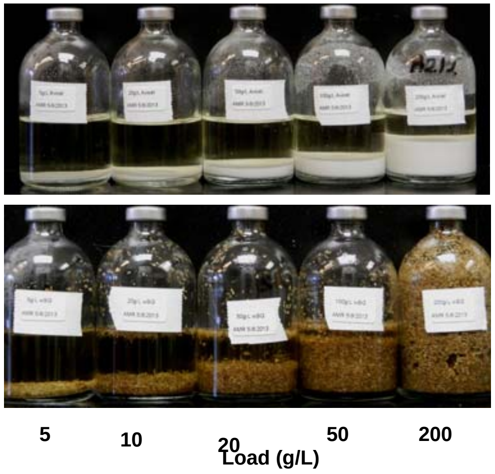
2015

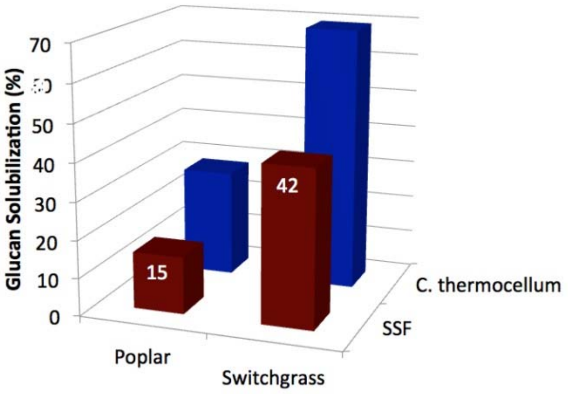
- Solubilization of plant cell walls more effective when mediated by cellulolytic microbes
- Multiple new structures and models for cellulosomes along with chimeric cellulosomal enzymes
- New genetic tools for thermophilic cellulolytic microbes
- Improved ethanol yields in *C. thermocellum* and demonstrated ethanol production in *Caldicelluosiruptor*
- Production of advanced biofuels in thermophilic microbes



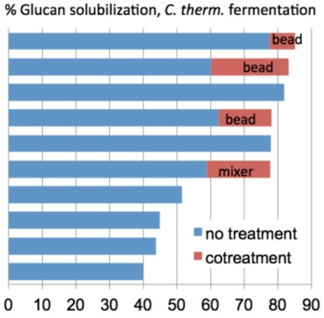
Early results: microbial solubilization of plant cell walls

- Larger than expected differences in solubilization effectiveness as a function of the choice of both feedstock and conversion system
- Ability of several anaerobic bacteria to solubilize cellulosic biomass without (or with minimal) conventional pretreatment
- *C. bescii* grows on high solids loadings (up to 200 g/L) of crystalline cellulose and non-pretreated switchgrass
- *C. bescii* solubilizes all components of non-pretreated switchgrass (cellulose/hemicellulose/lignin) at 78°C
- Better than 74% cellulose conversion of five grassy feedstocks by *C.thermocellum* with minimal pretreatment





	Max Particle Size (mm)		Feedstock Growth Stage		
	.08	~1-2	mid	late	washed
Switchgrass		✓	✓		
Corn Stover		✓		✓	
Corn Cob		✓		✓	
Switchgrass		✓	✓		✓
Winter Rye		✓	✓		
Bagasse*		✓		✓	
Winter Rye		✓		✓	
Poplar	✓				✓
Switchgrass	✓			✓	
Poplar	✓				



Cellulase architecture and mechanisms

Significance

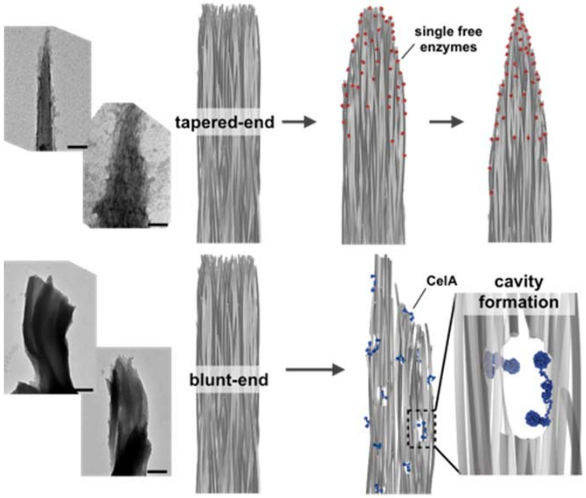
The different modes of action of CelA and Cel7A offer new possibilities for creating synergistic mixtures of biomass-attacking enzymes drawn from different organisms.

Outcome

- The activity of CelA on Avicel is sevenfold higher than the common exo/endo cellulase standard mixture, Cel7A and Cel5A.
- CelA appears to be both surface active and active in excavating into the substrate creating cavities and additional surface area for attack, whereas Cel7A is apparently strictly surface active and creates tapered ends on Avicel.

BESC Synergy

- Draws on deep NREL expertise in fungal cellulases, hydrolysis, and imaging.
- CelA identified as a key enzyme by Caldi groups at UGA and NCSU based on sequencing from JGI and transcriptomics and proteomics at NCSU and ORNL.



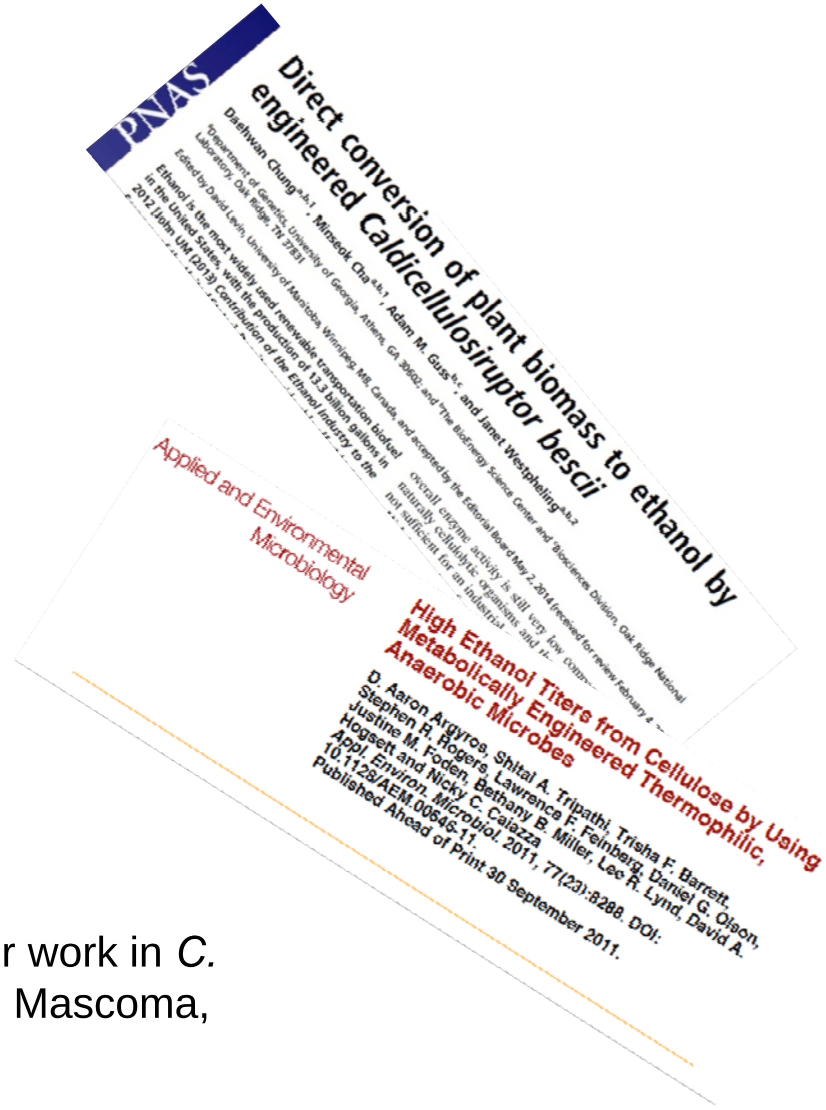
TEM micrographs of partially digested small Avicel PH101 particles. Those digested to ~60% conversion by a Ctec2 cellulase mixture (primarily comprising *T. reesei* Cel7A) display tip sharpening on one end. Particles digested to approximately ~65% conversion with CelA display slightly narrowed, tapered, blunt ends, as well as pits of various sizes on the surface. All scale bars are 500 nm.

Brunecky, R., *et al.*, *Science* 342:6165, 2013.

Early results: genetic systems for thermophiles

BESC reported successful DNA transformation of both *C. thermocellum* and *C. bescii*:

- Used methylation with an endogenous unique *α*-class N4-Cytosine methyltransferase to overcome major barrier to DNA transformation in *C. bescii.*
- Strategy for *C. thermocellum* employed counter selections developed from the native *C. thermocellum hpt* gene and the *T. saccharolyticum tdk* gene; was used to delete the genes for both lactate dehydrogenase (Ldh) and phosphotransacetylase (Pta).



PNAS

Direct conversion of plant biomass to ethanol by engineered *Caldicellulosiruptor bescii*

Daehwan Chung[a,b,1], Minseok Cha[a,b,1], Adam M. Guss[b,c], and Janet Westpheling[a,b,2]

[a]Department of Genetics, University of Georgia, Athens, GA 30602; and [b]The BioEnergy Science Center and [c]Biosciences Division, Oak Ridge National Laboratory, Oak Ridge, TN 37831

Edited by David Levin, University of Manitoba, Winnipeg, MB, Canada, and accepted by the Editorial Board May 2, 2014 (received for review February 4, 2

Ethanol is the most widely used renewable transportation biofuel in the United States, with the production of 13.3 billion gallons in 2012 [John UM (2013) Contribution of the Ethanol Industry to the

overall enzyme activity is still very low comp
naturally cellulolytic organisms and th
not sufficient for an industrial

Applied and Environmental Microbiology

High Ethanol Titers from Cellulose by Using Metabolically Engineered Thermophilic, Anaerobic Microbes

D. Aaron Argyros, Shital A. Tripathi, Trisha F. Barrett, Stephen R. Rogers, Lawrence F. Feinberg, Daniel G. Olson, Justine M. Foden, Bethany B. Miller, Lee R. Lynd, David A. Hogsett and Nicky C. Caiazza
Appl. Environ. Microbiol. 2011, 77(23):8288. DOI: 10.1128/AEM.00646-11.
Published Ahead of Print 30 September 2011.

BESC Synergy

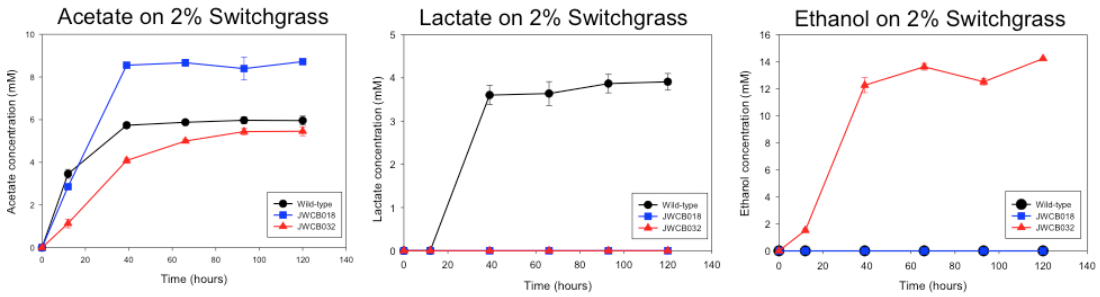
- Significant lessons learned were applied from prior work in *C. thermocellum* and *T. saccharolyticum* (Dartmouth, Mascoma, ORNL).
- The methods are being transferred into BESC partner labs at ORNL and NCSU for metabolic engineering for the direct conversion of lignocellulose to ethanol or butanol.

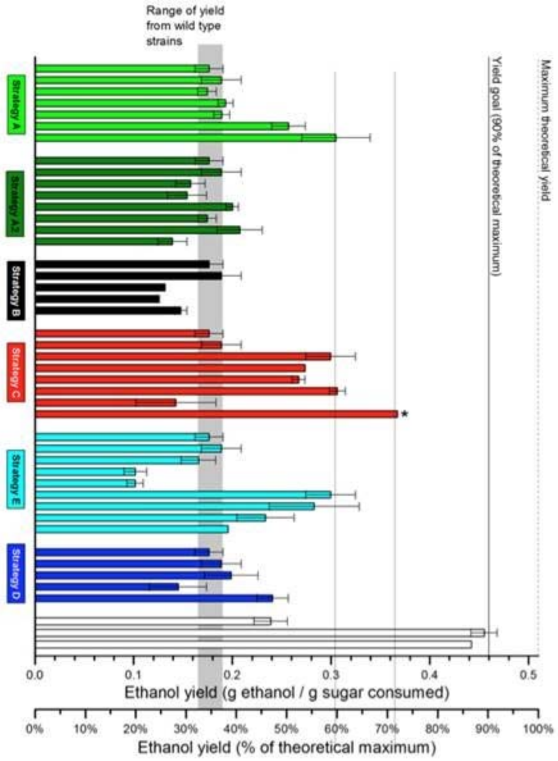
Building on efforts to increase biofuel yield in BESC CBP thermophiles

U.S. DEPARTMENT OF ENERGY | Office of Science

Outcome

- *C. thermocellum:*
  - Several engineering strategies have improved ethanol yield.
  - HydG was the best targeted mutation.
  - Several of the evolved strains share a point mutation in the *adhE* alcohol dehydrogenase.
- *C. bescii:*
  - Also based on incorporation of *adhE* gene;
  - 15 mM ethanol levels;
  - Acetate, lactate and ethanol production when grown on cellobiose, Avicel and switchgrass.



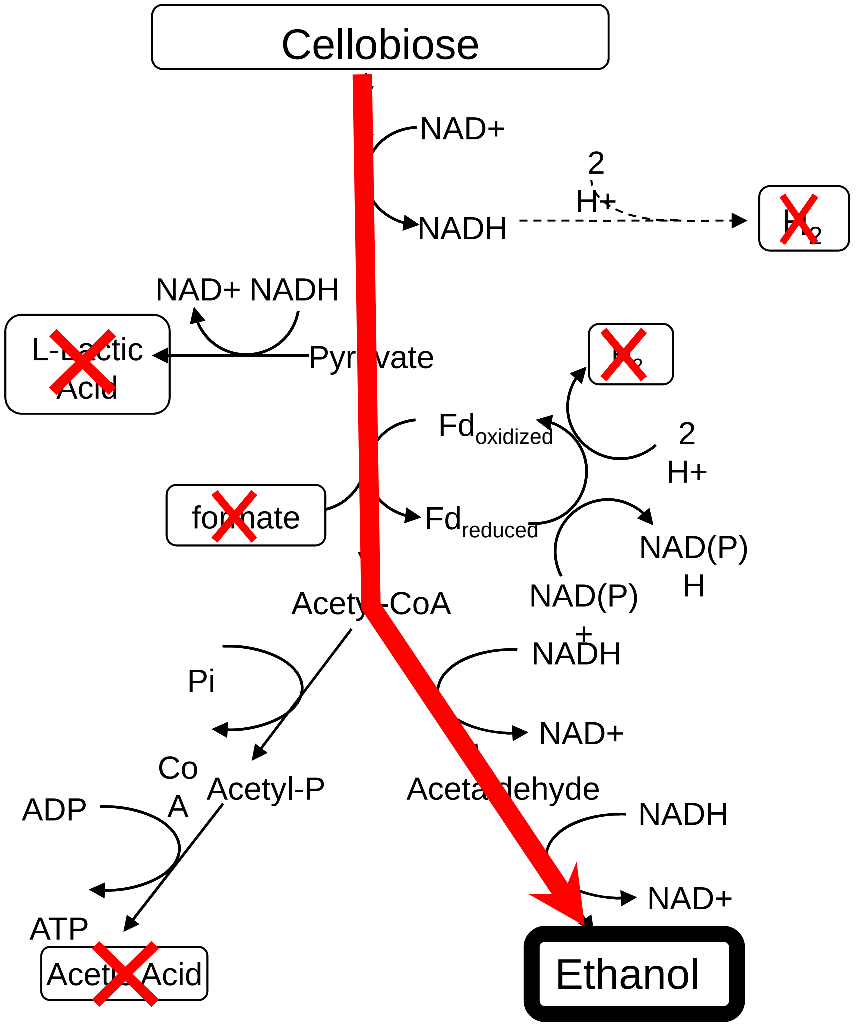


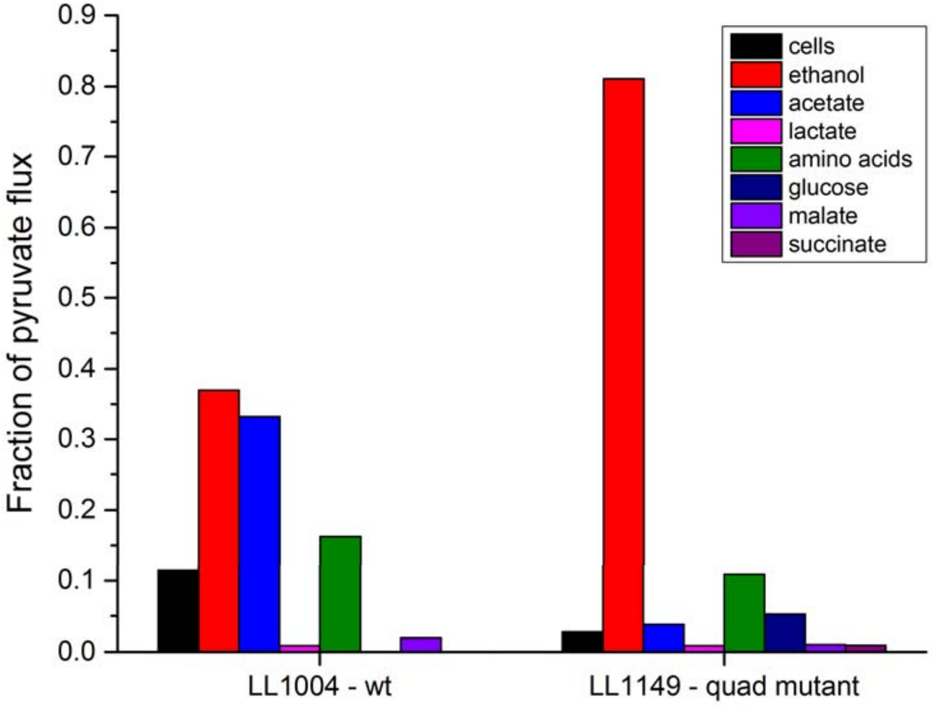
BESC Synergy

Shared strategies among Dartmouth, ORNL,UGA

BESC BioEnergy Science Center

Highest *C. thermocellum* ethanol yield to date: "quad mutant" (*pfl*$^-$ *hydG*$^-$ *ldh*$^-$ *pta-ack*$^-$)





Ethanol yield 81% of theoretical

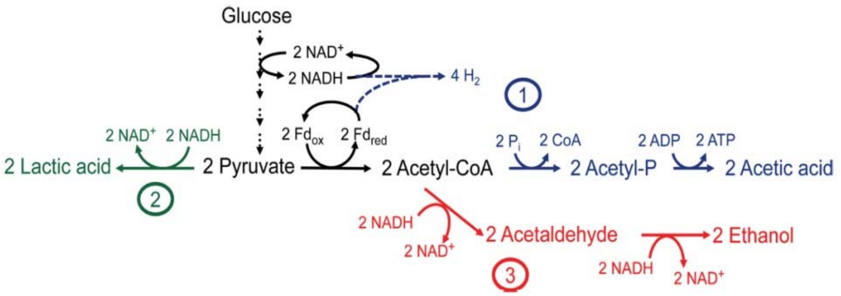
However strain grows very slowly – 10 days to consume 5 g/L cellobiose (lean medium) - secretes pyruvate, valine, alanine

Biswas *et al*., submitted

Conversion of switchgrass to biofuel using engineered *Caldicellulosiruptor bescii*

Significance

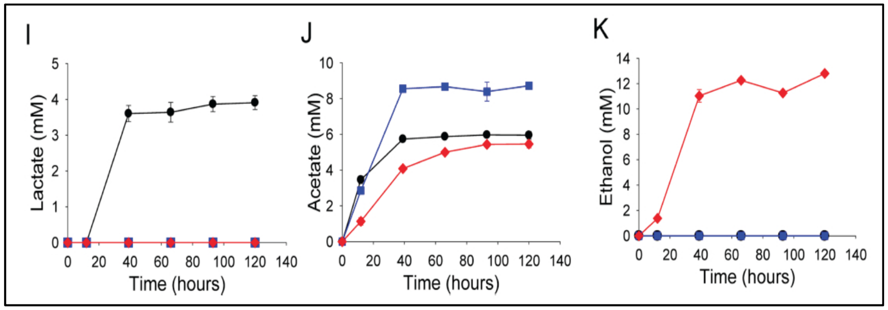
- Direct conversion of plant biomass to a fuel without pretreatment.
- First demonstration of metabolic engineering of an extreme thermophilic organism for conversion of lignocellulosic biomass to a liquid fuel.



Overview of *C. bescii* fermentative pathways for bioconversion of hexose sugars. Pathway 3 (red) is a new pathway resulting from heterologous expression of the *C. thermocellum adhE* gene to synthesize 2 moles of ethanol per mole of glucose.

Outcome

- The ldh^-, $AdhE^+$ strain redirected most of the carbon flux to ethanol, with 12.8mM ethanol produced from non-pretreated switchgrass (compared to 14.8mM from cellobiose).
- Switchgrass was neither chemically nor thermally pretreated prior to digestion.



Lactate, acetate and ethanol production by wild-type (I), ldh^- (J), and ldh^- $AdhE^+$ (K) strains of C *bescii* grown on non-pretreated switchgrass

Chung, *et al*., *PNAS* 111(24), 8931-8936, 2014.

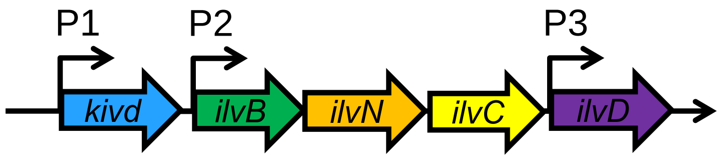
BESC Synergy

- Led by UGA with genetic engineering and fermentation, fermentation advice and HPLC at ORNL
- Draws on shared standard switchgrass (Noble) previous characterization (NREL) and ORNL fermentation protocols using pretreated materials

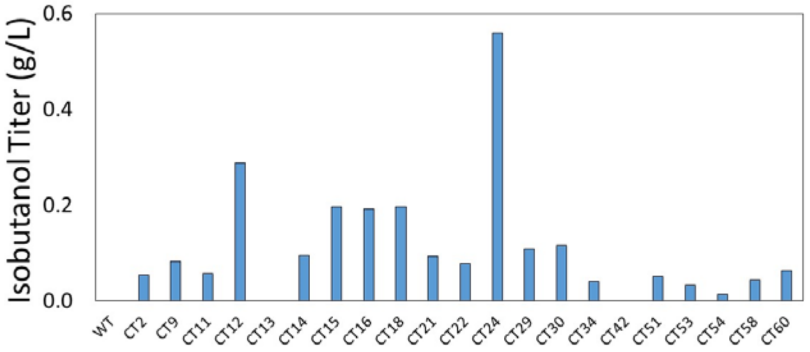
Isobutanol production from cellulose in *C. thermocellum*

120 plasmids with different promoters constructed

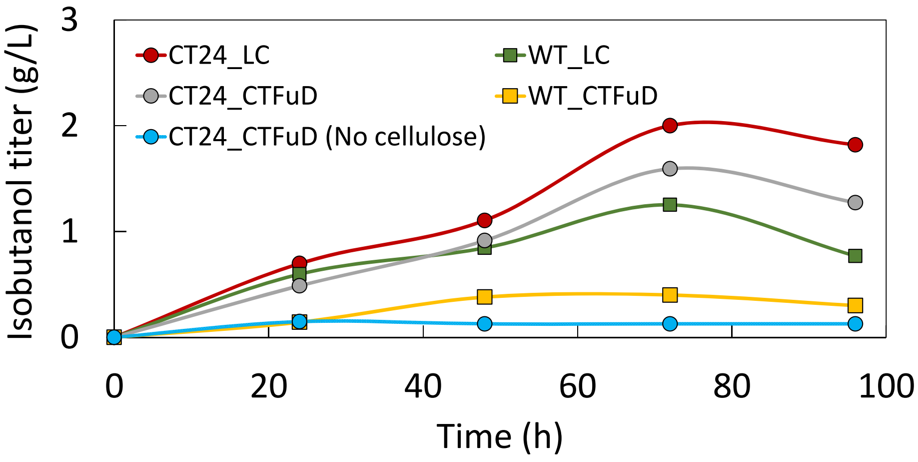
40 verified and tested in *C. thermocellum*



Isobutanol production in CTFuD medium (0.45% YE) during 24 h



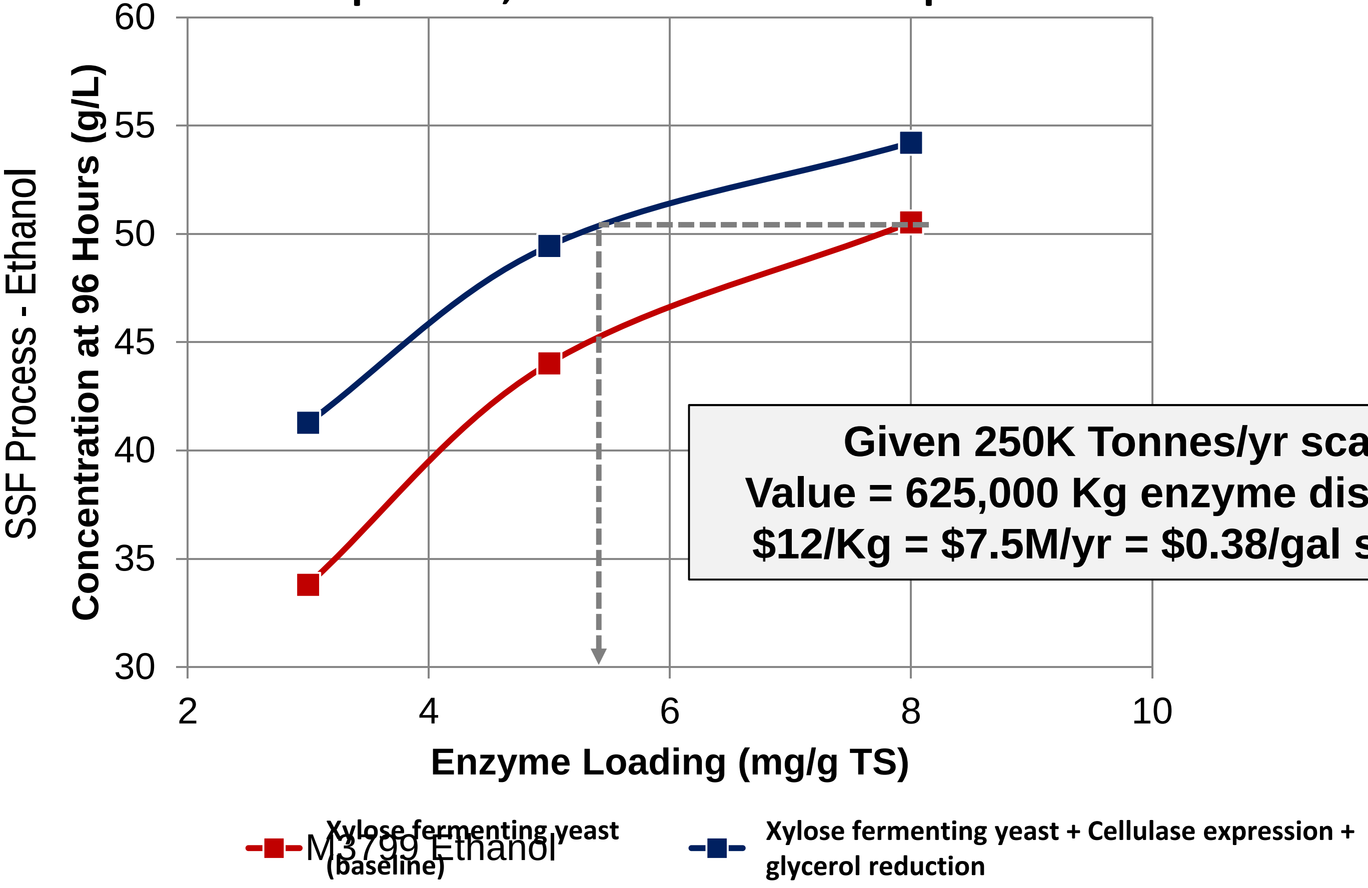
Isobutanol production in LC medium and CTFuD medium during 96 h



Achieved 2 g/L of isobutanol in minimal medium (LC medium)

Including cellulase expression and glycerol reduction in the C56X yeast

8-10% Yield Improvement or 2.5 mg/g TS cellulase displacement already demonstrated in an SSF process; transferable to HHF process



Given 250K Tonnes/yr scale:
Value = 625,000 Kg enzyme displaced:
$12/Kg = $7.5M/yr = $0.38/gal savings

Enabling technology: then and now

2007

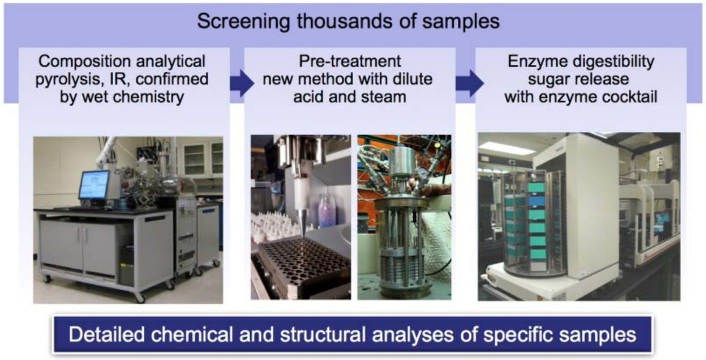
- Gold standard for biomass composition was NREL method requiring ~5 g per sample and over two weeks' effort
- Submicron analyses for catalytic imaging and chemical signatures of biomass were not established
- Limited polysaccharide epitope antibodies were just beginning to be used for imaging
- Very large biological datasets were genomic and primarily used in medical screening fields, not in bioenergy

2015

- High-throughput pipeline for analysis of composition and sugar release for thousands of samples per year
- Glycome profiling now medium throughput and validated for analyses of cell wall structure and automated
- Establishment of experimental and computational protocols protocols for high-performance proteomics
- Use of computational modeling to increase understanding of lignin biosynthesis and polymerization
- Integration of modified feedstocks with engineered microbes
- Metabolomics analysis coupled with genetic information to identify metabolite-gene associations

Development and utilization or sample analysis pipelines

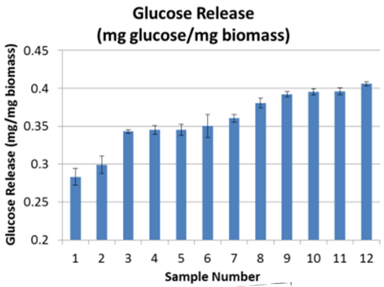
- High-throughput (1000s per month):
  - ➢ lignin composition
  - ➢ S/G ratio
  - ➢ glucose/xylose release
- Medium-throughput (100s):
  - ➢ sugar composition assay (new)
  - ➢ glycome profiling
  - ➢ bioconversion assay
- Low-throughput (10s)
  - ➢ high resolution image analysis
  - ➢ NMR (multiple modalities)
  - ➢ cellulose crystallinity
  - ➢ accessibility
- Over 65 techniques available
  - ➢ see BESC characterization handbook



Screening thousands of samples

Composition analytical pyrolysis, IR, confirmed by wet chemistry → Pre-treatment new method with dilute acid and steam → Enzyme digestibility sugar release with enzyme cocktail

Detailed chemical and structural analyses of specific samples



Glucose Release (mg glucose/mg biomass)

Bioenergy Science Center (BESC)
Biomass Characterization Technique Reference
Volume 2
May 2011

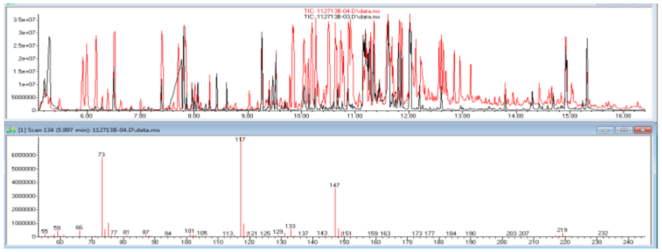
Support for identification and characterization of TOP Lines

- High-throughput cell wall chemistry and recalcitrance screening (NREL)
  - Assay lignin content, S/G ratio and sugar release identifying low recalcitrance lines and prioritizing deeper characterization work.
  - ~ 30,000 samples planned for BESC Years 6-8.
- Biomass analytical workflow (UGA, NREL, UCR)
  - Use chemical, immunological, enzymatic, and spectroscopic methods to identify changes in biomass structure (TOP selection).
  - Provide predictive knowledge to relate recalcitrance and structure.
- Whole-cell wall NMR with emphasis on lignin structure (UTK/ORNL, UGA)
  - Whole-cell ionic liquid 1D and 2D NMR to characterize the structure of lignin and hemicelluloses and correlate changes with lowered recalcitrance (TOP selection).
- Small-scale cell wall compositional assays (NREL, ORNL)
  - Analyze major and minor sugars as well as other hydrolyzate products (e.g., acetate, furfural, and HMF).
  - Screen TOP candidates for fermentation inhibitors.
- Screening for cellulose crystallinity, accessibility, and degree of polymerization (DP) (UTK/ORNL)
  - Monitor and understand changes to cellulose in transgenic lines and correlate changes to reductions in recalcitrance (TOP selection).

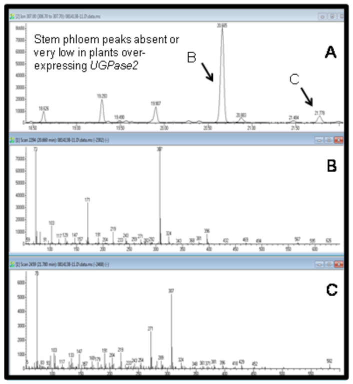
Capabilities of ’omics

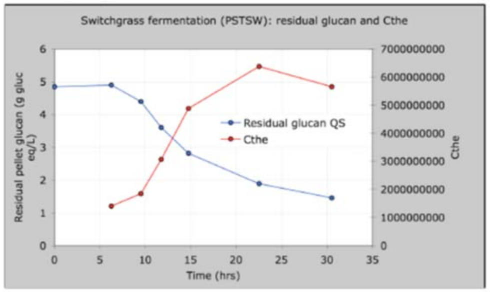
Outcome

- Metabolomic profiling identifies growth inhibitors , fuel production pathways, and microbial intracellular redox metabolites:
  - ➢ Characterized and quantified aromatic metabolites and known microbial inhibitors released and generated by *Thermoanaerobacter saccharolyticum* degrading corn cob biomass
- Metabolomic profiling of transgenic plants versus controls:
  - ➢ Completed the GC-MS analysis of >60 transgenic lines of *Populus deltoides, Panicum virgatum*, and *Arabidopsis thaliana.*
- A proteomics-based assay measuring *C. thermocellum* cell density grown on biomass substrates:
  - ➢ Applied to real-world samples (right), tMIT specifically and accurately tracks cell density across cultures that utilize both simple (cellobiose) and complex (switchgrass) carbon sources.



Thermoanaerobacter saccharolyticum - corn cob supernatant (red) vs T0 control (black)





BESC Synergy

- ORNL ’omics expertise applied to Dartmouth, Noble, UGA and ORNL samples
- Drivers for improved methods in microbial and redox measurements came from FA2

Combining modified switchgrass with engineered *C. thermocellum* improves yield

Significance

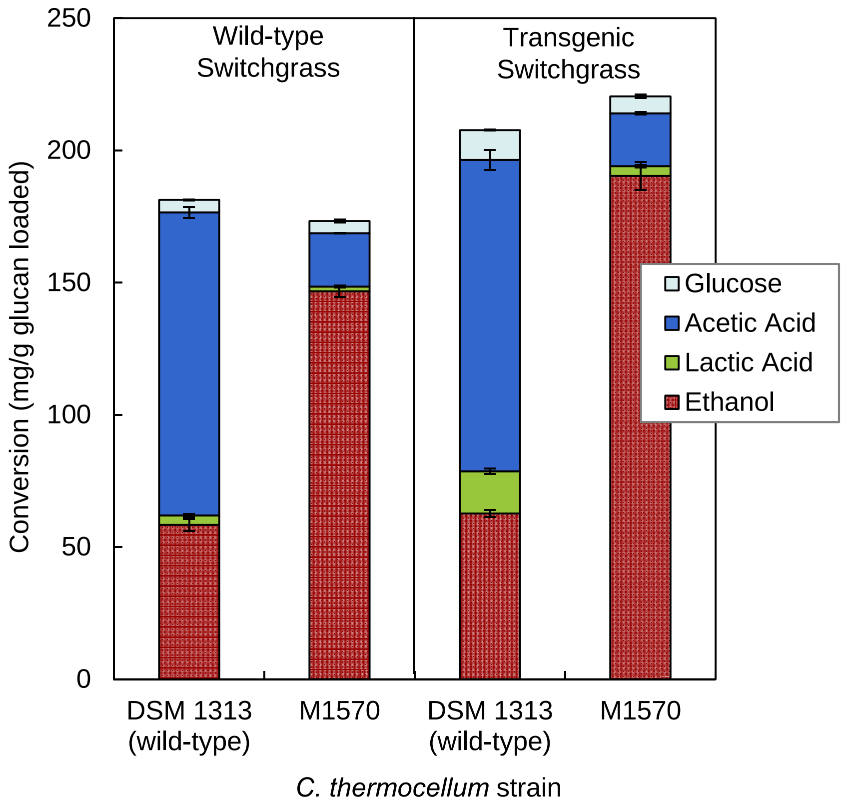
- First report of use of a microbe engineered to produce increased amounts of a biofuel on a bioenergy feedstock modified for the same purpose. Results demonstrate the potential additive advantages from combining a modified feedstock with an engineered consolidated bioprocessing microorganism.

Outcome

- Fermentation of the modified switchgrass by *C. thermocellum* mutant M1570 had superior conversion relative to the wild-type control switchgrass line with an increase in conversion of approximately 20%.
- Ethanol was the primary product, accounting for 90% of the total metabolites with conversion of 0.19 g ethanol/g glucan loaded and 0.27 g liberated.

BESC Synergy

- Combines elements from across BESC: COMT switchgrass (Noble), *C. thermocellum* (Mascoma) with ORNL expertise.



Conversion (mg/g glucan loaded) for *C. thermocellum* mutant M1570 and wild-type DSM 1313 strains on both transgenic (T1-3-TG) and wild-type (T1-3-WT) switchgrass, which were pretreated with dilute acid. The standard deviation is from the average of triplicate buffered serum bottle fermentations.

Yee *et al.*, *Biotechnology for Biofuels* 7:75, 2014.

Community outreach in bioenergy science education is now self sustaining

Farming for Fuels lessons reach thousands of students through hands-on science activities

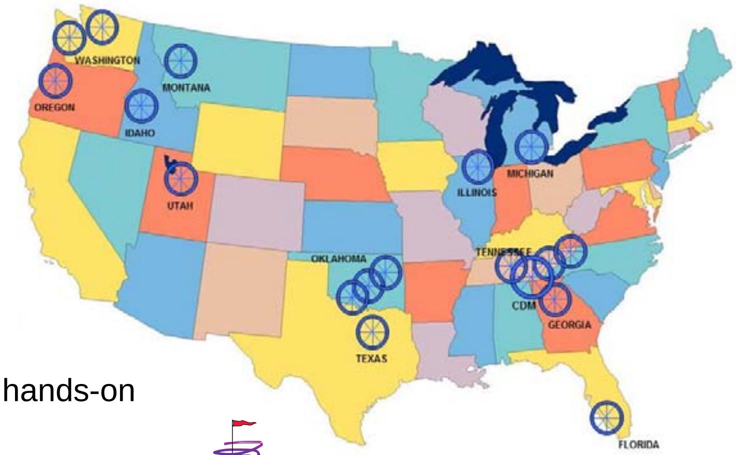
- BESC in collaboration with the Creative Discovery Museum (CDM) in Chattanooga, Tennessee, developed hands-on lesson plans for students in 4th, 5th and 6th grades.
- *Farming for Fuel* lessons educate students about the carbon cycle, lignocellulosic biomass as substrate for the production of biofuels and the technical and economic obstacles to a bio-based fuel economy.

Science Night events reach thousands of families

- In the last 2 years, 170 Science Nights were presented nation-wide reaching more than 25,000 students, parents and teachers.

“Hub and Spoke” model allows economical outreach national outreach using partnering with regional science centers and museums. Over six years, the outreach program has steadily expanded from Chattanooga across Tennessee to currently active hubs in Georgia, Texas, Michigan, Illinois, Florida, Oklahoma, Idaho, Montana, Washington, Oregon and Utah.

- ➢ A marker of self-sustaining success is that now 75% of the support for the hands-on activities now come from the schools, hubs, and other sources.
- ➢ This approach has allowed BESC to steadily increase hands-on science contacts to over 36,000 in the last year and over 116,000 students, parents, and teachers in the past six years.



CREATIVE DISCOVERY MUSEUM

BESC BioEnergy Science Center

BESC Active "Alumni"

BESC
BioEnergy Science Center

Where are they now?

Wen-Chi "Jacky" Chou
Post Doc Harvard University
formerly Research Assistant at University of Georgia

Adam Guss
Genetic and Metabolic Engineer at Oak Ridge National Laboratory
formerly Post Doc Dartmouth College

William "Bill" Adney
Director of Biofuels Research at RTI international
formerly Senior Scientist at National Renewable Energy Laboratory

Yanbin Yin
Assistant Professor at Northern Illinois University
formerly Assistant Research Scientist at The University of Georgia

Javier Izquierdo
Bioenergy Program Manager at RTI International
formerly Research Scientist at Dartmouth College

Jaclyn DeMartini
Scientist at Genencor
formerly PhD Graduate Student at The University of California - Riverside

Andrew Bordner
Assistant Professor of Pharmacology Mayo Clinic
formerly Post Doc University of Minnesota

Wen Zhou
Assistant Professor Michigan Technological University
formerly Post Doc at University of Georgia

Gancho Slavov
Lecturer at Aberystwyth University
formerly Researcher at University of West Virginia

Yu-San Liu
Chemist at Oso BioPharmaceuticals Manufacturing
formerly Researcher at National Renewable Energy Laboratory

Anastasia Aksenova
Graduate Teaching Assistant at UT-Knoxville
formerly Student Intern at ORNL

Shihui Yang
Scientist at the National Renewable Energy Laboratory
formerly Post Doc at ORNL

Amy Van Fossen
Postdoctoral Research Assistant at Agrivida
formerly Postdoctoral Research Assistant at North Carolina State University

Joel Farkas
Post Doctoral Researcher at Ohio State University
formerly Graduate Student at The University of Georgia

Babu Raman
Senior Scientist at Dow AgroSciences
formerly Postdoctoral Research Associate at Oak Ridge National Laboratory

Bingqiang Liu
Lecturer at School of Mathematics Shandong University
formerly Student at University of Georgia

Fengfeng Zhou
Professor at Shenzhen Institute of Advanced Technology
formerly Research Scientist at The University of Georgia

Poulomi Sannigrahi
Associate Scientist at Phillips 66
formerly Postdoctoral Researcher at Georgia Institute of Technology

Zhiwu Wang
Assistant Professor at Ohio State University
formerly Post Doc at ORNL

Hongjia Li
Scientist at Genencor
formerly Graduate Student at The University of California - Riverside

Adriane Lochner
Public Relations Associate at College Hill in Munich Germany
formerly Post Doc at Oak Ridge National Laboratory

Kimberly Hunt
Instructor at Georgia Southern University
formerly Post Doc at UGA

J. D. Hogan
Post Doc at Boston University
formerly Graduate Research Assistant at UGA

Yongchao Li
Post Doc University of Oklahoma
formerly Post Doc at ORNL

Scott Hamilton-Brehm
Post Doc at Desert Research Institute
formerly Post Doc at Oak Ridge National Laboratory

Marcus Foston
Assistant Professor at Washington University in St. Louis
formerly Research Scientist at Georgia Institute of Technology

Chris Hubbell
Research Scientist at Alcon Laboratories
formerly Postdoctoral Fellow at Georgia Institute of Technology

Future BESC Alumni

Heidi Hau
Ecolab
formerly Scientist at Mascoma

Inci Ozdemir
Senior Scientist at Pfizer
formerly Research Assistant at North Carolina State University

Rajeev Kumar
Assistant Research Engineer at University of California – Riverside
formerly Post Doc at Dartmouth College

Wendy Higashide
Research Scientist at Easel Biotechnologies
formerly Research Assistant at UCLA

Heather McKenzie
Assistant Professor at The University of British Columbia
formerly a PhD Graduate Student at The University of California - Riverside

Douwe van der Veen
Technology Associate at Protospace / Fablab Utrecht
formerly Postdoctoral Fellow at Dartmouth College

Elements of BESC’s value system

- Focus on impactful recalcitrance science
- A high-functioning team of world-class scientists
- Emphasis on integration and collaboration
- Close connection to industry increasing potential of impact
- Acceleration of research and technology outcomes
- Growing core of well-trained young research staff

Thank you!

U.S. DEPARTMENT OF ENERGY | Office of Science

BESC
BioEnergy Science Center


One more thing...

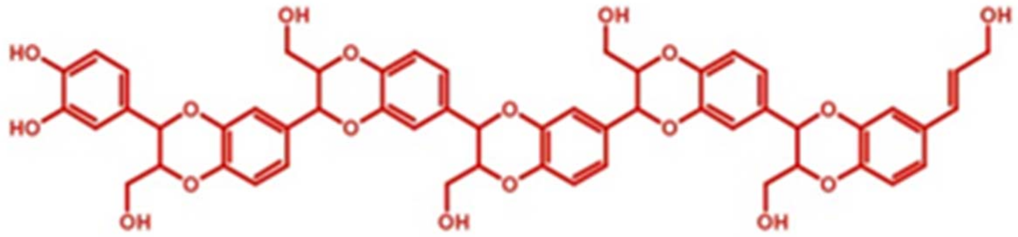
Reduced distortion with reinforced materials

ABS

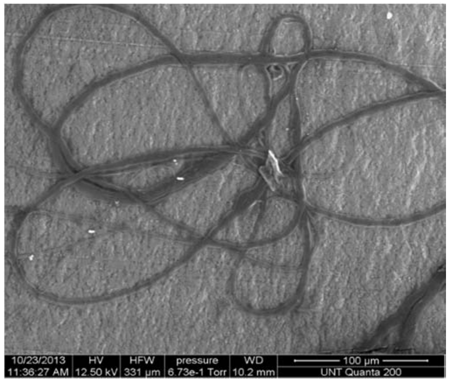
15% CF-ABS

C-lignin shows promise as a potential co-product for biorefining

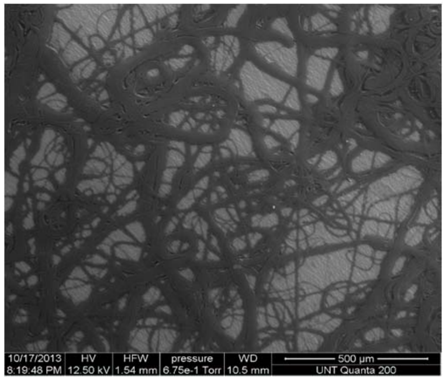
May be easier to extract from plant cell walls. Replacing G/S lignin with C-lignin might lead to reduced recalcitrance of biomass. **Linear nature** makes this molecule better suited than classical lignin for manufacture of **carbon fibers** and bioplastics

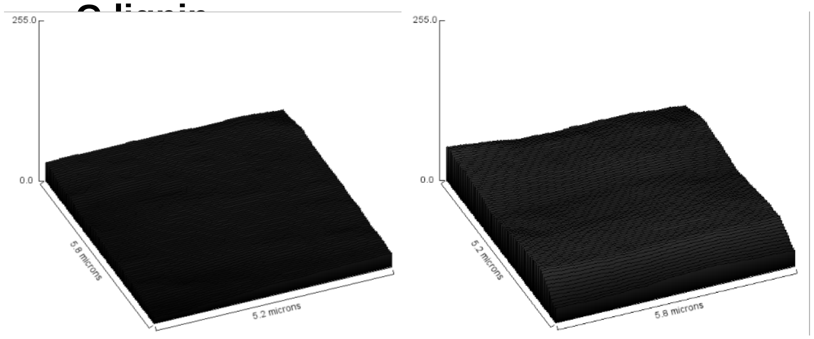


C-lignin



Kraft lignin





Nar et al., unpublished results