Office of Biological and Environmental Research

Biological Systems Science Division Update

Todd Anderson, Ph.D.
Director, Biological Systems Science Division,
Department of Energy, Office of Biological &
Environmental Research

October 27, 2016

Programmatic Activities

Completed Reviews/Activities

- Pre-application screening for the re-competition of the Bioenergy Research Centers (BRCs) FOA
- Panel review and selection for the Plant Feedstocks for Bioenergy FOA
- Panel review and selection for the Microbial Carbon Cycling FOA
- Bioimaging project review and panel review of new projects
- Panel review of Harvard-led project on Integrated Genome, Ribosome, and Proteome Engineering
- Three National Laboratory Science Focus Area (SFA) reviews
- Workshop on "*Technologies for Characterizing Molecular and Cellular Systems Relevant to Bioenergy and Environment*"

Upcoming Reviews/Activities

- Year 9 annual progress review of the Bioenergy Research Centers (Nov)
- Panel review of applications submitted to the re-competition FOA for the Bioenergy Research Centers (Dec)
- Planning for the upcoming Genomic Science PI meeting (Feb)
- Initial plans BSSD COV (Jul)

Bioenergy Research Centers Open Competition

Sustainability

**Feedstock Development**

Biomass Deconstruction

Conversion biofuels & bioproducts

- Team-based, multi-institutional, cross-disciplinary, integrated systems research centers.
- Total requested budget of $89 million per year.
- Center annual budgets may range from $12.5M to $30M for up to 5 years.
- Open competition for domestic organizations as prime applicant; open to any organization as subawardee.
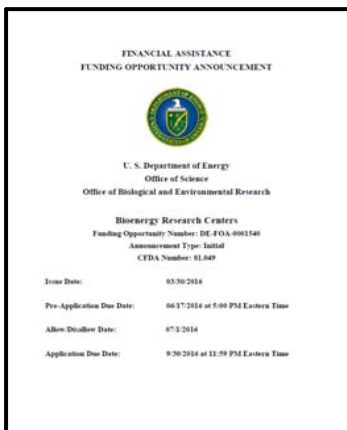
FOA (DE-FOA-0001540)
Posted - 3/30/2016
**Applications due - 9/30/2016**
FOA Closed
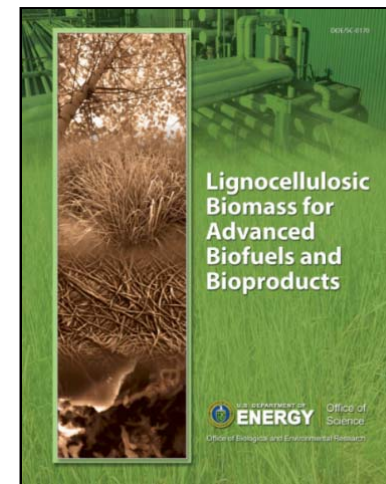
New Awards for FY 2016

Plant Feedstock Genomics for Bioenergy: A Joint Research Funding Opportunity Announcement USDA, DOE (DE-FOA-0001444)

➤ *Continues research on plant responses to pathogens*

➤ *Includes a focus on oil seed crops*

7 new projects ($7.8M over 3 yrs; 2 funded by USDA)

Plant Feedstock Genomics for Bioenergy Joint Awards 2006–2016
genomicscience.energy.gov/research/DOEUSDA/

- **E. Sacks (UIUC)** – *"Introgression of Novel Disease Resistance Genes from Miscanthus into Energycane"*

- **S. Edme (USDA)** – *"Genetics and genomics of pathogen resistance in switchgrass"*

- **A. Alonso (Ohio State Univ.)** *"Development of Resources and tools improving oil content and quality in Pennycress "*

- **J. Brown (Univ. Idaho)** – *"Developing Non-food Grade Brassica Biofuel Feedstock Cultivars with High Yield, Oil Content, and Oil Quality Suitable for Low Input Production Dryland Systems"*

- **C. Lu (Montana St.)** – *"Systems Biology to Improve Camelina Seed and Oil Quality Traits"*

- **J. Dyer (USDA)** – *"Genomics and Phenomics to Identify Yield and Drought Tolerance Alleles for Improvement of Camelina as a Biofuel Crop"*

- **D. Funnell-Harris (USDA)** – *"Resistance to stalk pathogens for bioenergy sorghum"*

New Awards for FY 2016
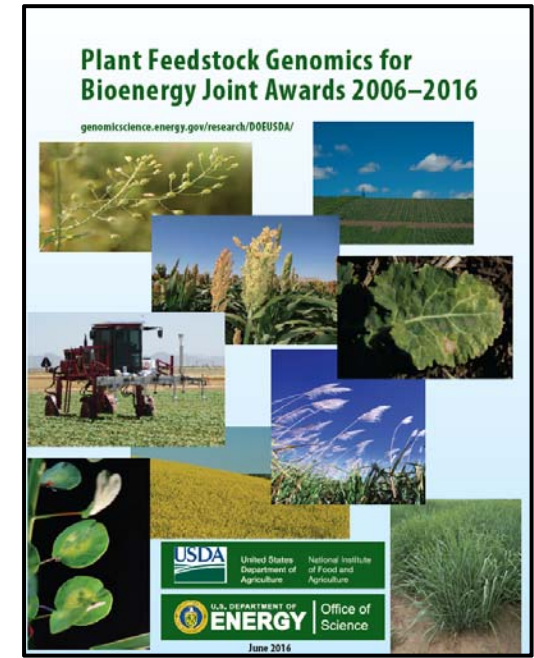
Carbon Cycle Research

Systems Biology Enabled Research on the Roles of Microbial Communities in Carbon Cycle Processes (DE-FOA-0001458)

➢ *Research to understand the functioning of microbial communities in a wide range of environments and environmental conditions*
➢ *Research to inform larger scale ecosystem function*
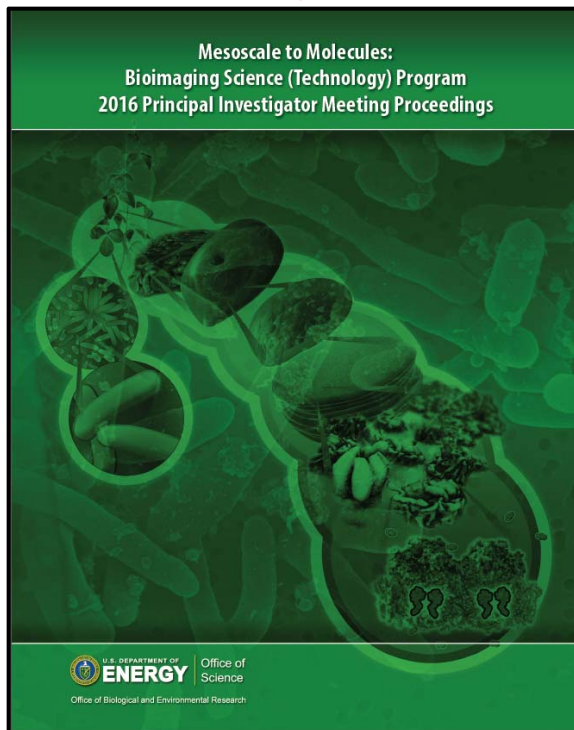
9 new projects ($30M over 3yrs)

- **V. Orphan (CalTech)** – "*Identification and ecophysiological understanding of new microbial players, processes, and multi-scale interactions in the global methane cycle*"
- **M. Firestone (UCBerk)** – "*Directing traffic in the rhizosphere: how phage and fauna shape the flow and fate of root carbon through microbial pathways*"
- **S. Allison (UC-Irvine)** – "*A trait-based framework for linking microbial communities with carbon transformations under precipitation change*"
- **T. Bruns (UCBerk)** – "*Determination of the Roles of Pyrophilous microbes in the Breakdown and Sequestration of Pyrolyzed Forms of SOM*"
- **B. Hungate (N. Ariz)** – "*Scaling the Microbial Ecology of Soil Carbon*"
- **V. Rich (Ohio State)** – "*Illuminating the pathways to carbon liberation: a systems approach to characterizing the consequential unknowns of carbon transformation and loss from thawing permafrost peatlands*"
- **K. DeAngelis (UMass)** – "*The "Who" and "How" of Microbial Control over Soil Carbon Dynamics: a Multi -omics, Stable Isotope Probing, and Modeling Approach*"
- **D. Buckley (Cornell)** – "*Microbial metabolic dependency and its impacts on the soil carbon cycle*"
- **M. Lidstrom (Univ. Wash.)** – "Systems level understanding of interconnected methane and nitrate cycles using complex synthetic communities"
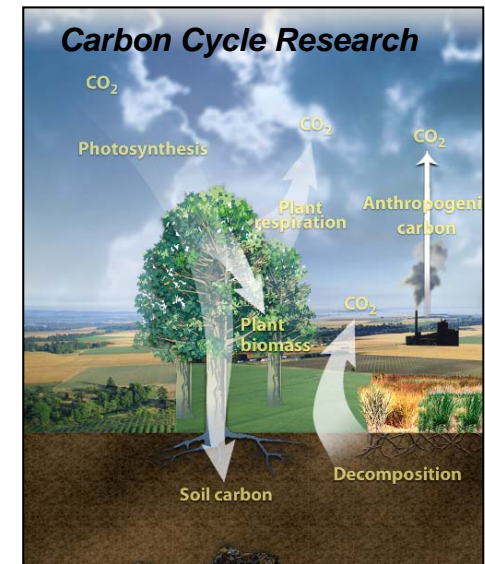
Bioimaging Technology Program

New imaging and measurement technologies to visualize metabolic processes governing phenotypic expression in plants and microbes

First PI meeting!

Mesoscale to Molecules:
Bioimaging Science (Technology) Program
2016 Principal Investigator Meeting Proceedings

Proceedings available at:
http://science.energy.gov/~/media/ber/pdf/community-resources/2016_Bioimaging_Technologies_PI_Meeting.pdf

DOE National Laboratories

- **Argonne National Laboratory,** _Kenneth Kemner_
- **Oak Ridge National Laboratory,** _Mitch Doktycz_
- **Pacific Northwest National Laboratory,** _James Evans_
- **SLAC Nat. Accelerator Laboratory,** _Soichi Wakatsuki_
- **AMES National Laboratory, Emily Smith**
- **Pacific Northwest National Laboratory, Scott Lea**

University Portfolio

- **Duke University,** _Tuan Vo-Dinh_
- **Iowa State University,** _Basil Nikolau_
- **Texas A&M University,** _Wayne Versaw_
- **University of Connecticut,** _Leslie Shor_
- **University of Missouri, Columbia,** _Gary Stacey_
- **University of North Carolina at Chapel Hill,** _Elizabeth Shank_
- **Washington University in St. Louis,** _David Fike_

New FOAs for FY 2017

Biosystems Design to Enable Next-Generation Biofuels and Bioproducts (DE-FOA-0001650)
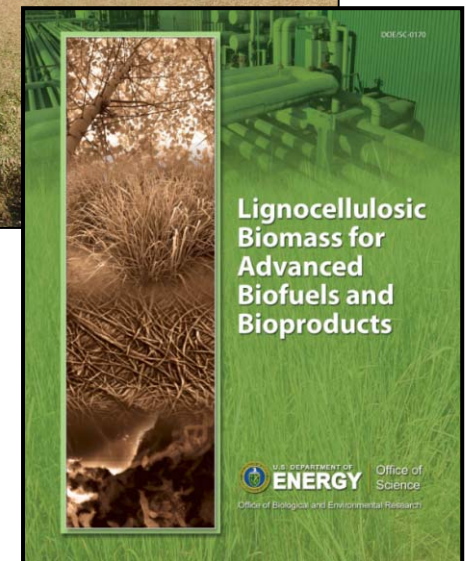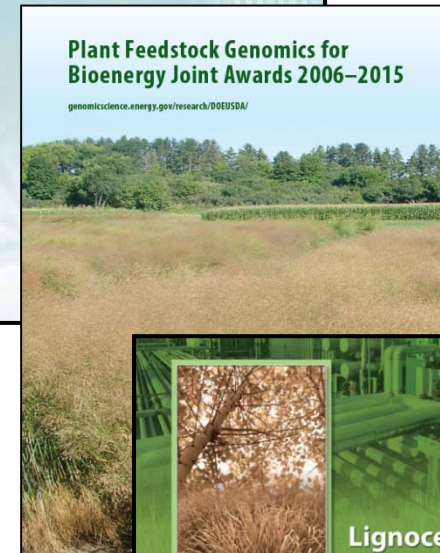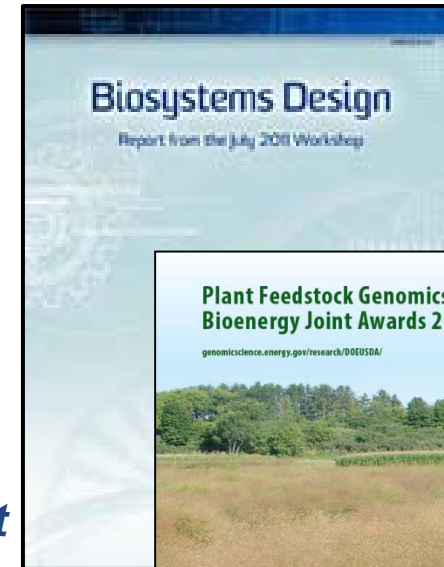**Posted** *10/12/16*
Pre-apps due 12/19/16
Full proposals due 3/20/17

Plant Feedstock Genomics for Bioenergy: A Joint Research Funding Opportunity Announcement USDA, DOE
(Pending)

Microbiome Research
(Pending Budget Resolution)

Department of Energy • Office of Science • Biological and Environmental Research

Strategic Planning: New Workshops

ASCR-BER Workshop on Exascale Computational Requirements
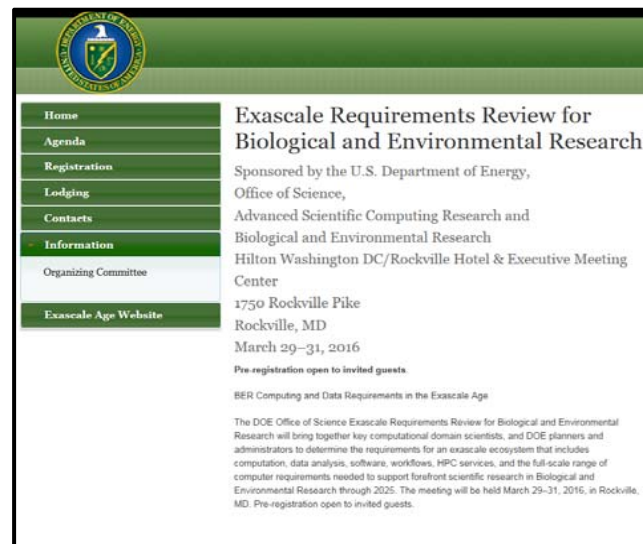
➢ March 29-31, 2016

➢ Hilton Washington DC/Rockville Hotel

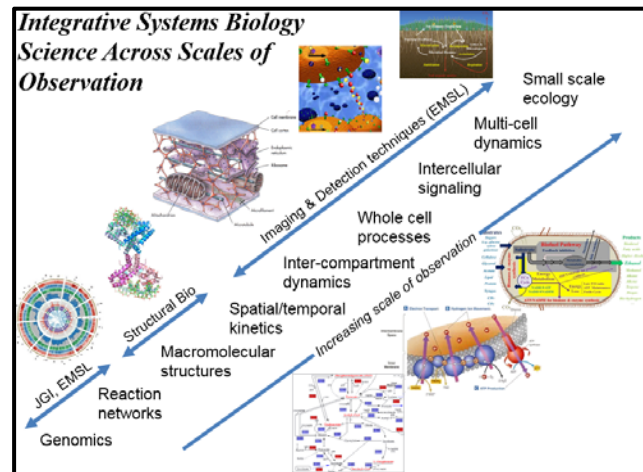*Dr. Jim Hack (ORNL) will provide an update.*

Workshop on Integrated Molecular Structure, Function and Dynamic Imaging Technologies

➢ Enabling *in situ* Quantitative Biology

➢ September 21-23, 2016

➢ Hilton Hotel, Rockville, MD

Dr. Amy Swain (BER) will provide an update

KBase
PREDICTIVE BIOLOGY
DOE Systems Biology Knowledgebase

http://www.kbase.us

The DOE Systems Biology Knowledgebase
A Community Resource for Predictive Biology

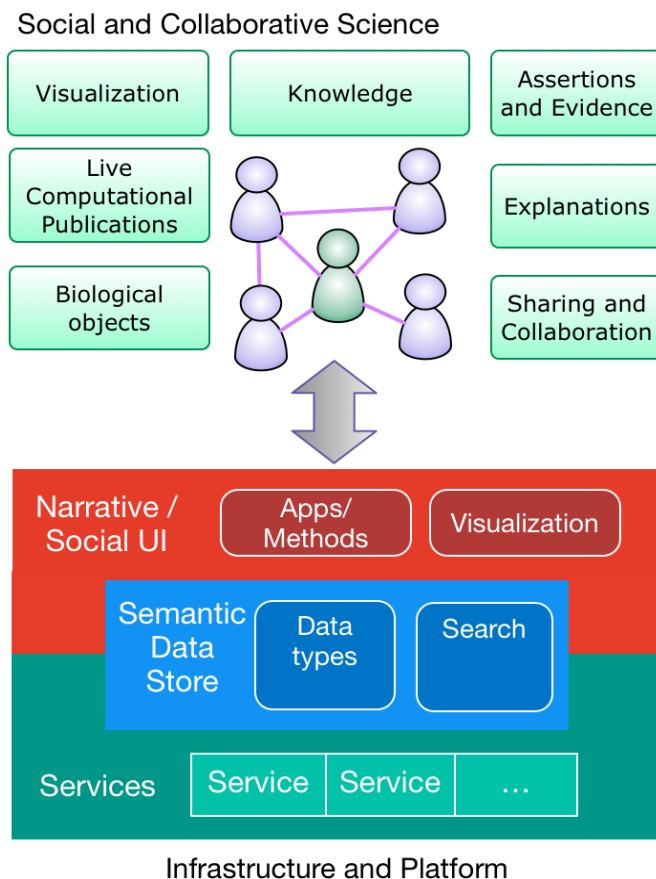**A socially-enabled data science platform for biology**

Open software and data platform for addressing the grand challenge of systems biology: **Predicting and designing biological function**

Unified system that integrates data and analytical tools for comparative functional genomics of **microbes, plants, and their communities**

Collaborative environment for **sharing methods and results** and placing those results in the context of knowledge in the field

Social and Collaborative Science

| Visualization | Knowledge | Assertions and Evidence |

| Live Computational Publications | | Explanations |

| Biological objects | | Sharing and Collaboration |

Narrative / Social UI — Apps/ Methods — Visualization

Semantic Data Store — Data types — Search

Services — Service — Service — …

Infrastructure and Platform

The KBase platform is designed to support the emergence of collaborative science and a knowledgebase for predictive biology

Department of Energy • Office of Science • Biological and Environmental Research
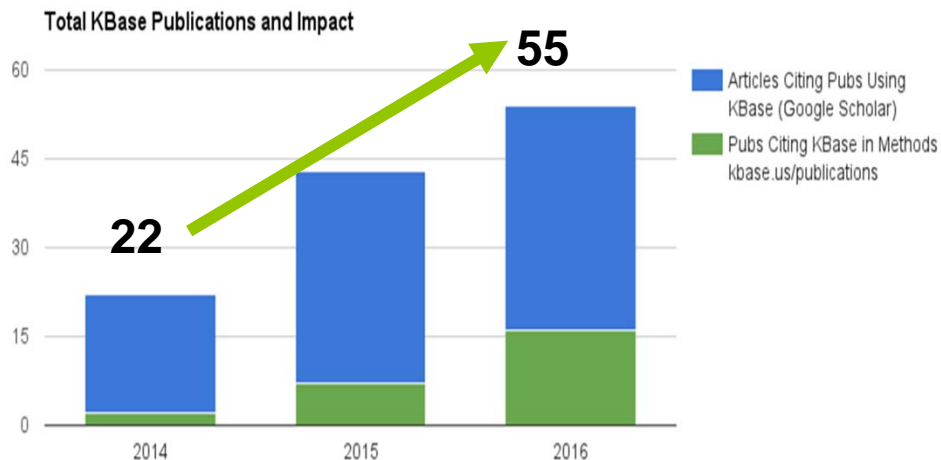
Utilization of KBase provides impactful S&T results and benefits to external user communities
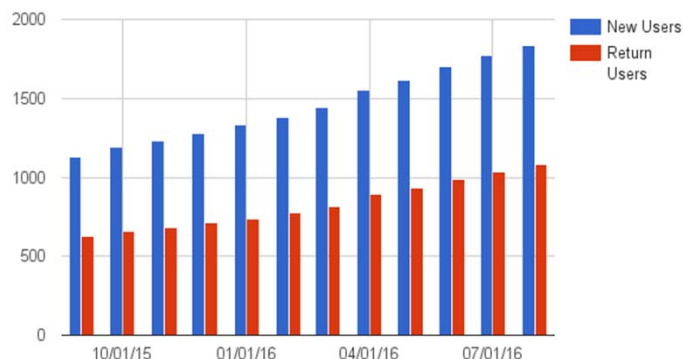
KBase impact is growing over time
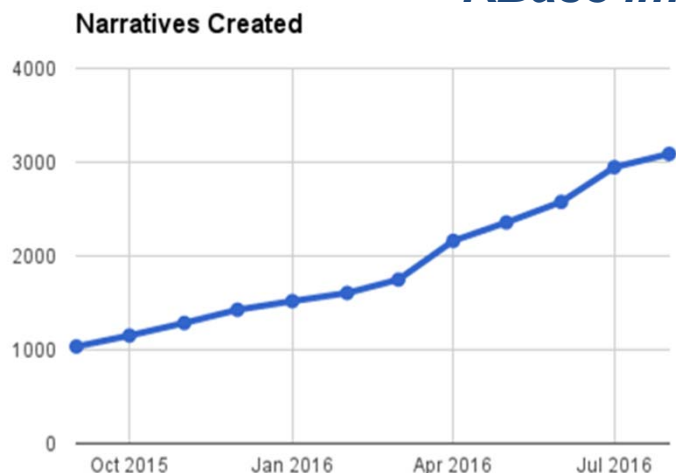
Growing number of Narratives (workflows) created by users

Increasing visibility in research literature

11 KBase workshops in 2016, 200+ attendees

400+ visitors to KBase booths

Department of Energy • Office of Science • Biological and Environmental Research

Microbial Community Metabolic Modeling: A Community Data-Driven Network Reconstruction

Objective: Develop models of microbial community metabolic networks to reliably predict interspecies metabolic interactions

Approach

➢ Reconstruction of single species and consortia metabolic models using the KBase platform
➢ Developed a binary consortium to model photoautotroph–heterotroph communities
➢ Used community-level data from consortia as critical input for community network reconstruction to predict interspecies metabolic interactions
➢ Simulations provided experimentally validated predictions of phototroph–heterotroph interactions

Results/Impact

➢ Coupled use of data and tools to implement a dynamic modelling approach in a reproducible workflow using KBase
➢ A new modeling strategy developed using community-level data as critical input for network reconstruction and multi-species dynamic modeling to directly predict interspecies metabolic interactions in a community, when axenic information is insufficient

Cover article, *Journal of Cellular Physiology*
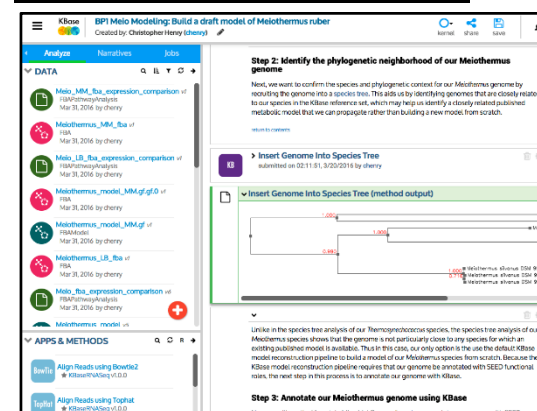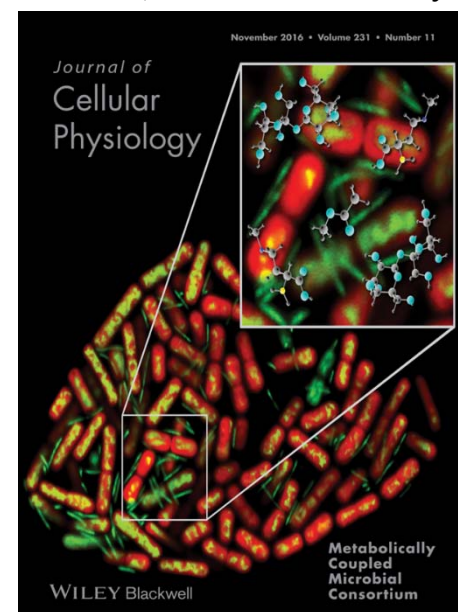
Reproducible Narrative available in KBase!

Henry, C. S., Bernstein, H. C., Weisenhorn, P., Taylor, R. C., Lee, J.-Y., Zucker, J. and Song, H.-S. (2016), Microbial Community Metabolic Modeling: A Community Data-Driven Network Reconstruction. **J. Cell. Physiol**. DOI: 10.1002/jcp.25428

BESC
BioEnergy Science Center

A bifunctional ammonia-lyase is critical in grass cell wall biosynthesis of lignin
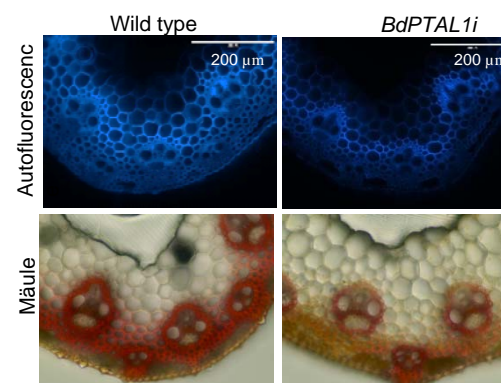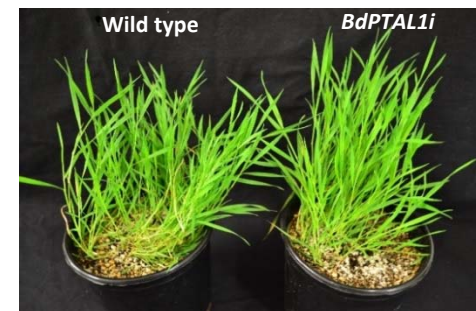

Objective: Identify biomarkers for lignin synthesis in grasses

Approach

- Using phylogenetic, transcriptomic and *in vitro* biochemical analyses, a single cytosolic bifunctional ammonia-lyase (PTAL) was identified the model grass species *Brachypodium distachyon*.

- ^{13}C stable isotopic labelling along with *BdPTAL1*-down-regulation in transgenic plants showed that the TAL activity can provide nearly half of the total lignin deposited in grasses, with a significant preference for S-lignin.

Results/Impact

- PTAL function is linked to the characteristic features of grass cell walls (high S/G ratio and presence of wall-bound coumaric acid).

- Organization of lignin synthesis enzymes is located in one or more metabolons. It remains to be determined whether the parallel pathways to lignin from L-phenylalanine and L-tyrosine feature separate enzymes or common enzymes used in different metabolic channels.

- TAL is an new target for metabolic engineering of lignin content in order to reduce recalcitrance. Because S/G ratio is directly proportional to TAL/PAL activity ratio, it may also be possible to engineer high S lignins through over-expression of PTAL.
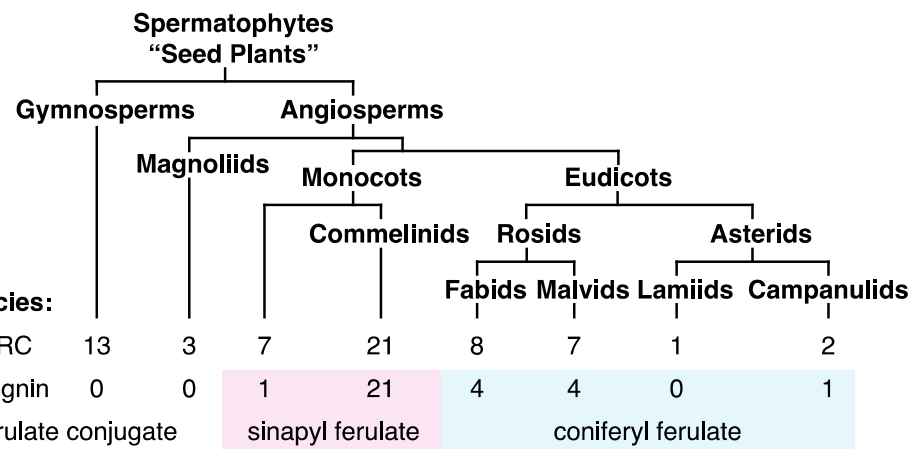
Barros, J., Serrani-Yarce, J.C., Chen, F., Baxter, D., Venables, B.J. and Dixon, R.A., "Role of bifunctional ammonia-lyase in grass cell wall biosynthesis," ***Nature Plants,*** 2016, 2:16050

Plants have convergently evolved to use "zips" (chemically labile ester linkages) in their lignin polymers

Objective: Survey a set of plants representing the spermatophytes or "seed plants" for the presence of monolignol (ML) ferulate conjugate ester linkages ("zips") within the lignin polymers of these plants

Approach

➢ Detect presence and/or incorporation of chemically labile ML-ferulate-derived ester bonds (zips) in lignin isolated from various gymnosperm and angiosperm species
➢ Conduct phylogenetic analysis of BAHD acyl transferases

	Spermatophytes "Seed Plants"						
	Gymnosperms	Angiosperms					
		Magnoliids	Monocots		Eudicots		
				Commelinids	Rosids		Asterids
					Fabids	Malvids	Lamiids	Campanulids
Number of species:							
Analyzed by DFRC	13	3	7	21	8	7	1	2
Positive for zip-lignin	0	0	1	21	4	4	0	1
Dominant ML-ferulate conjugate			sinapyl ferulate		coniferyl ferulate			

Result/Impacts

➢ Previous work demonstrating zip-lignin in commercially relevant poplar engineered with an exotic Ferulate Monolignol Transferase (FMT) revealed that wild-type poplar trees already have low, but detectable, zip-levels in their lignins.
➢ All gymnosperms tested showed no evidence of zip-lignin, whereas low zip-levels were present in the isolated lignins of many, but not all, angiosperms; sinapyl ferulate dominated in monocots, whereas coniferyl ferulate dominated in dicots.
➢ Identification of plant lines with increased zip-lignin content, likely able to be more easily and economically deconstructed, could provide superior sources of biomass for conversion to biofuels and bioproducts.

Karlen, S.D. *et al.* "Monolignol ferulate conjugates are naturally incorporated into plant lignins**." *Science Advances* **2**, e1600393 (2016) [DOI: 10.1126/sciadv.1600393].

Development of an E. coli strain for One-pot Biofuel Production from Ionic Liquid Pretreated Cellulose and Switchgrass

jbei
Joint BioEnergy Institute

Objective: Engineer IL tolerant microorganisms for biofuel production

Approach

- An IL-tolerant production host was equipped with IL-tolerant cellulases to produce the advanced biofuel precursor d-limonene from cellulose and hydrolysate in the presence of IL
- Spontaneous resistance of *E. coli* to an a widely used IL was determined to be due to a knock out phenotype in a single gene, *rcdA*.
- This strain was able to produce limonene using saccharified IL-pretreated hydrolysate as carbon source
- Equipping this strain with an IL-tolerant cellulase (J29) allowed production of limonene from unsaccharified cellulose in the presence of 100 mM [C2C1im][OAc]
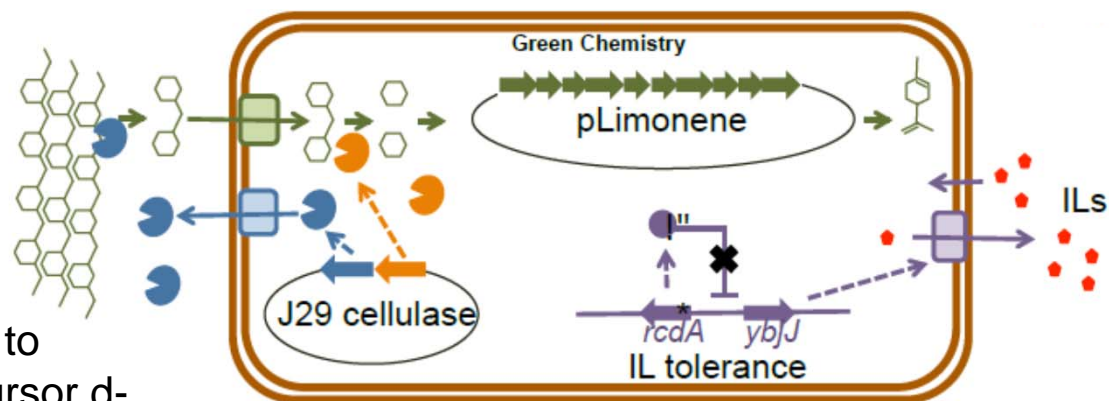
Results/Impact

- This work represents the necessary next step in the development of one-pot biofuel production processes in the presence of ionic liquids

M. Frederix *et al.* (2016). "Development of an E. coli strain for one-pot biofuel production from ionic liquid pretreated cellulose and switchgrass," ***Green Chemistry***, 18; 4189-4197 DOI: 10.1039/C6GC00642F

Structural Basis of Stereospecificity in Enzymatic Cleavage of Lignin Bonds

GREAT LAKES BIOENERGY RESEARCH CENTER

jbei Joint BioEnergy Institute

Joint Pubs!

Objective: Determine the structural basis for stereo-specificity of bacterial enzymes involved in β-aryl ether bond cleavage of lignin

Approach: Solve crystal structures and conduct biochemical analyses for LigE and LigF protein variants

Result/Impacts

➢ LigE and LigF possess dramatically different structural arrangements within the monomers and different dimer interfaces; as a result, the substrate binding surfaces of these two enzymes are on opposite faces, hence introducing stereospecificity.

➢ As lignin is the most abundant aromatic polymer in nature, this collaborative study[1] by GLBRC, JBEI, and others, combined with a second study,[2] informs broader lignin valorization efforts that will ultimately enable the development of efficient pathways for the conversion of lignin into renewable aromatics with applications in advanced biofuels and chemicals.
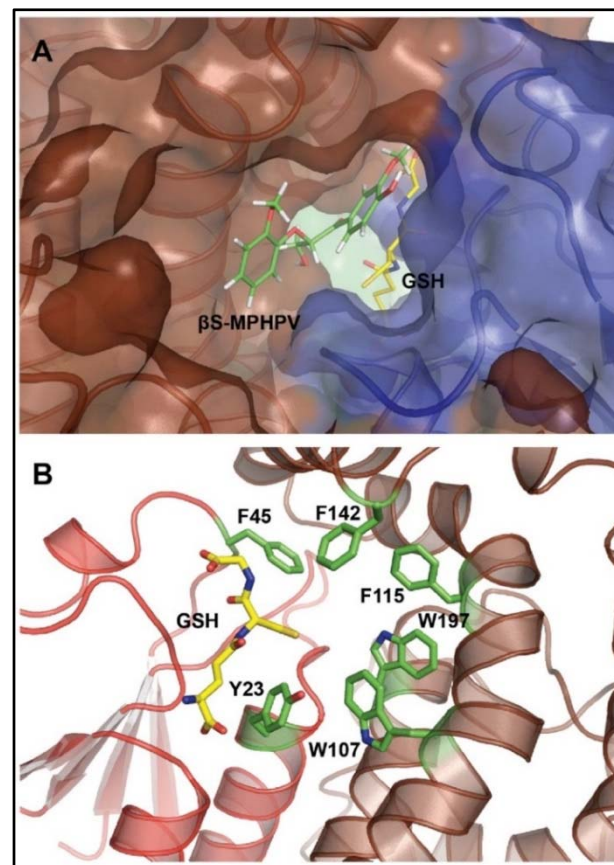
[1] Helmich KE, et al. 2016. *Structural basis of stereospecificity in the bacterial enzymatic cleavage of b-aryl ether bonds in lignin.* **The Journal of Biological Chemistry**, doi:10.1074/jbc.M115.69430.

[2] Pereira, JH, et al. 2016. *Structural and biochemical characterization of the early and late enzymes in the lignin b-aryl ether cleavage pathway from Sphingobium sp SYK-6.* **The Journal of Biological Chemistry**, doi:10.1074/jbc.M115.700427.

Brown Rot Two-Step: How Brown Rot Fungi Efficiently Degrade Wood

Early Career Awardee!

Objective: Examine the genetic "connectomics" of how brown rot fungi decompose wood efficiently, despite having lost many of the genes critical for plant cell wall deconstruction.

Approach

- Grew brown rot in one direction on thin wood sections, then cut the wood wafers into sections representing a different time point of wood decay.
- Each section was analyzed for gene expression, assayed for relevant enzyme activity and imaged using confocal and fluorescence microscopy.

Results/Impact

- Sections early in colonization showed upregulation of oxidative pathways and expression of plant cell wall-swelling genes both potentially weakening the structural integrity of wood.
- Only later time points found brown rot fungi expressing cellulose degradation enzymes.
- This unique fungal "pre-treatment" strategy where oxidants and enzymes are separated by time, instead of space, could lead to improved biotechnology approaches for efficient and cost effective conversion of woody plant materials into biofuels or bioproducts.

Recoding the E. coli genome

Objective: Synthetize a recoded bacterial genome that uses 57 of the 64 natural codons, allowing the reassignment of 7 codons to non-standard amino acids to develop novel protein functions.

Approach

A recoded and reduced *E. coli* genome was designed and synthesized in 87 fragments spanning 50 kb each. Testing of 55 of these fragments that contain 63% of the genome and 52% of essential genes showed that there were only minimal effects on growth and transcription levels. Redesigning the few recoded genes that caused fitness impairment, restored fitness of the engineered strain.

Results/Impact

➢ A recoded bacterial genome was designed and construction and testing was initiated.
➢ The feasibility of high level recoding of microbial organisms to confer new functionality such as the development of new bioproducts was demonstrated.
➢ Genome-wide engineering provides new knowledge on the fundamental principles that drive biological systems, such as protein synthesis.
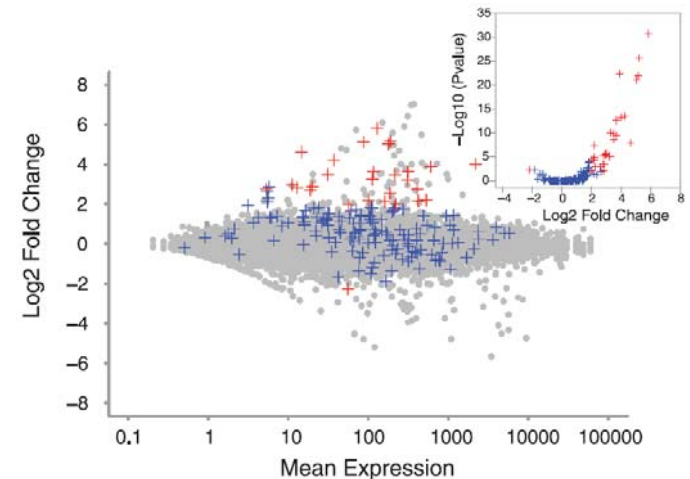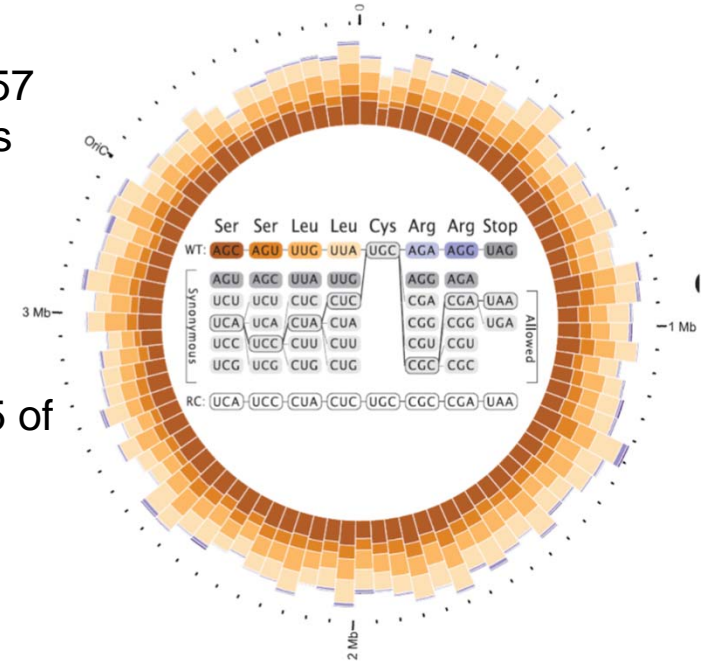
Ostrov *et al.*, "Design, synthesis, and testing toward a 57-codon genome." *Science* **353**, 819 (2016).

Neutrons Provide Insight into the Structure of Cellulose Synthase Complex for Potential Biofuel Applications

OAK RIDGE | CENTER FOR
National Laboratory | STRUCTURAL MOLECULAR BIOLOGY

Objective: Understanding the mechanism of cellulose synthesis in plants for potential biofuel applications

Approach

➢ ORNL researchers solved a low resolution structure of the catalytic domain of the cellulose synthase complex (CSC).

➢ Neutron and X-ray scattering together with computational techniques provide the first experimental evidence to reveal a self-assembly of "hexamer of trimers".

Results/Impacts

➢ Data support that the CSC enzyme is a "hexamer of trimers" model, that synthesizes an 18-chain cellulose microfibril as its fundamental product.

➢ This study advances understanding of the plant cellulose synthase (CESA), which forms a multimeric complex (CSC) responsible for synthesis of cellulose chains and their assembly into microfibrils
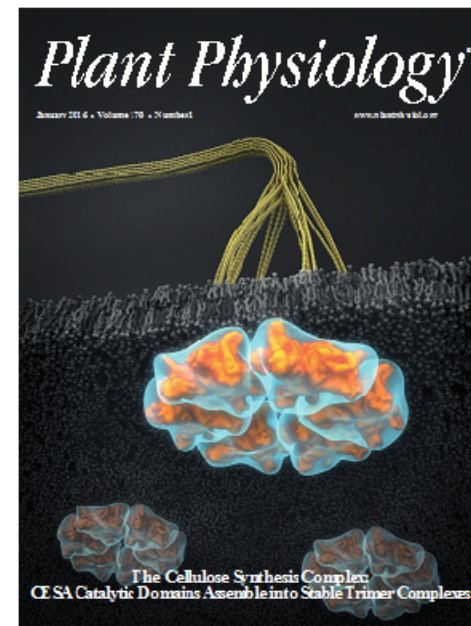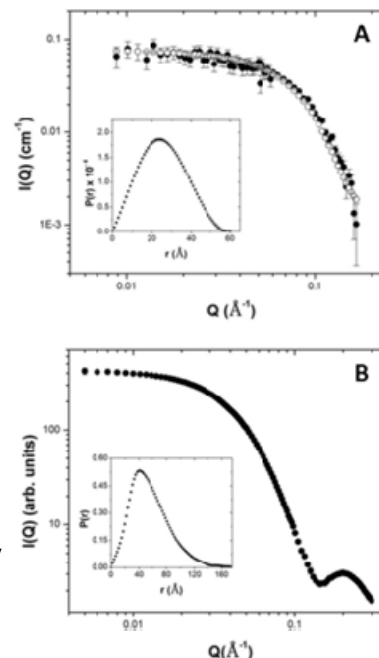
Left: SANS and SAXS profiles of CatD monomers and trimers. Right: Plant Physiology *cover art showing CSC. Ab initio models of catalytic domain of CESA calculated from scattering data are represented by semi-transparent grey surface envelopes, superimposed with the computational atomic models in orange*

V.G. Vandavasi, D.K. Putnam, Q. Zhang, L. Petridis, W.T. Heller, B.T. Nixon, C.H. Haigler, U. Kalluri, L. Coates, P. Langan, J. Smith, J. Meiler, and H.M. O'Neill. A Structural Study of CESA1 Catalytic Domain of Arabidopsis Cellulose Synthesis Complex: Evidence for CESA Trimers1. ***Plant Physiology*** (2016) 170.1, 123–135.

JGI — **JOINT GENOME INSTITUTE**
A DOE OFFICE OF SCIENCE USER FACILITY
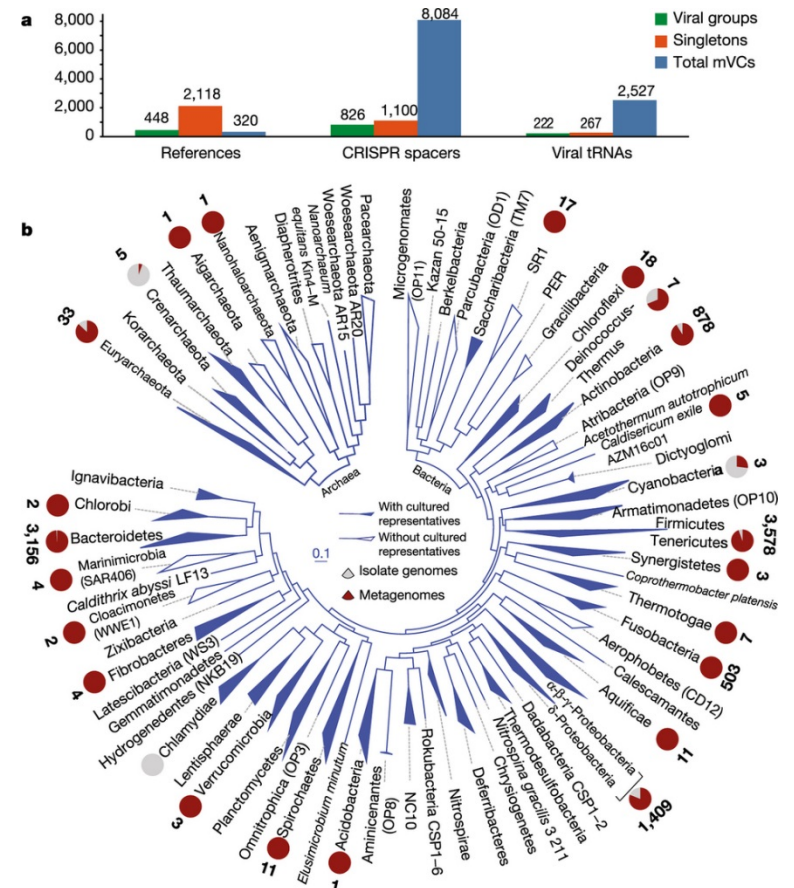
Uncovering Earth's Virome

Objective: Identify viral signatures in metagenomics analyses from a wide range of environments

Approach

➤ A combination of metagenomics sequencing and new computational analysis techniques enabled an unprecedented survey of viruses detectable in the environment

➤ Analysis of ~5Tb of metagenomic sequence data from 3,042 globally diverse environmental samples detected more than 125,000 partial viral genomes

➤ This increases the number of known viral genes by 16-fold

➤ Using CRISPR sequences, viral genes could be related to microbial hosts

Results/Impacts

➤ Doubles the number of microbial phyla known to be infected by viruses and identified viruses that can infect organisms from other phyla.

➤ Highlights the global diversity of viruses and provide detailed insight into viral habitat and distribution

Paez-Espino et. al. (2016) "*Uncovering Earth's Virome*", **Nature** 536, pp.425-430, 25 August, DOI:10.1038/nature19094

Selected Publications in 2016:

- *Uncovering Earth's virome*, **Nature**
- *SAR11 bacteria linked to ocean anoxia and nitrogen loss,* **Nature**
- *The genome of the seagrass Zostera marina reveals angiosperm adaptation to the sea,* **Nature**
- *Sequencing wild and cultivated cassava reveals genetic diversity,* **Nature Biotechnology**
- *Global metagenomic survey reveals a new bacterial candidate phylum in geothermal springs,* **Nature Communications**
- *Integrative genomic mining for enzyme function to enable engineering of a non-natural biosynthetic pathway,* **Nature Communications**
- *Host phenotype and age shape the leaf and root microbiomes of a wild perennial plant,* **Nature Communications**
- *Expansion of signal transduction pathways in fungi by extensive genome duplication,* **Current Biology**
- *Facile recoding of selenocysteine in nature,* **Angewandte Chemie International Ed**
- *Visualizing in situ translational activity for identifying and sorting slow-growing archaeal-bacterial consortia,* **PNAS**
- *Comparative genomics of biotechnologically important yeasts,* **PNAS**

158 Total peer-reviewed publications (as of 9/21)
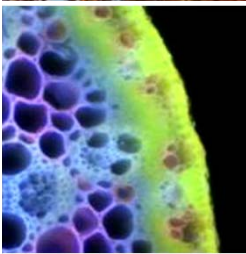
JGI Publication Highlights: http://jgi.doe.gov/news-publications/publications/
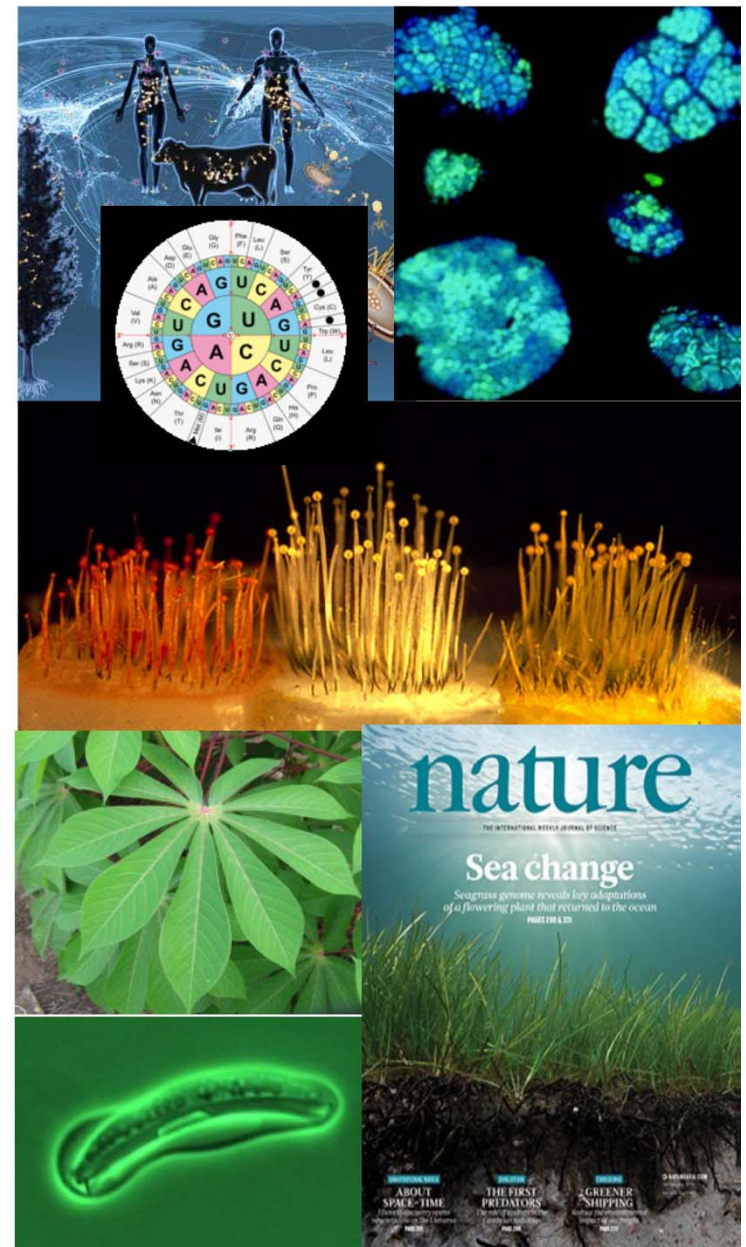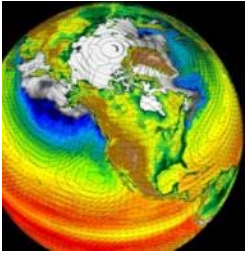
Systems science to meet DOE mission needs in bioenergy, climate and the environment.

http://science.energy.gov/ber

Thank you!

http://genomicscience.energy.gov