



U.S. DEPARTMENT OF
ENERGY

Office of
Science

Office of Biological and Environmental Research Biological Systems Science Division Update

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Energy, Office of Biological & Environmental Research

April 16, 2020

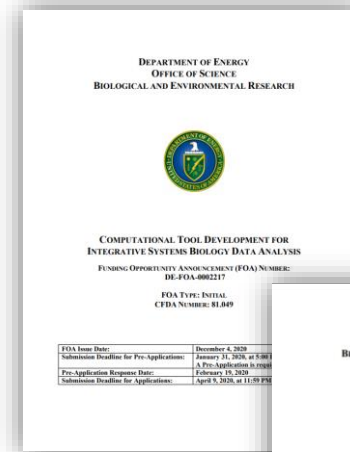
New Funding Opportunity Announcements for FY 2020

DE-FOA-002217

COMPUTATIONAL TOOL DEVELOPMENT FOR INTEGRATIVE SYSTEMS BIOLOGY DATA ANALYSIS

Proposals due: 4-16-2020

Extension to 4-30-2020 (case-by-case basis)

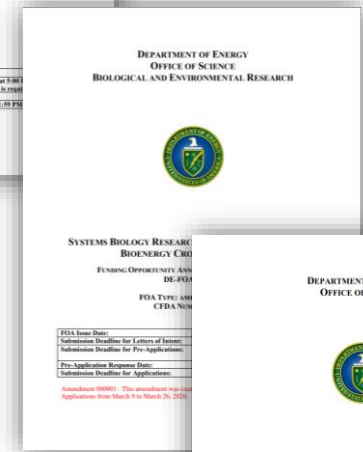


DE-FOA-002214

SYSTEMS BIOLOGY RESEARCH TO ADVANCE SUSTAINABLE BIOENERGY CROP DEVELOPMENT

Proposals due: 4-2-2020

Extension to 4-16-2020 (case-by-case basis)

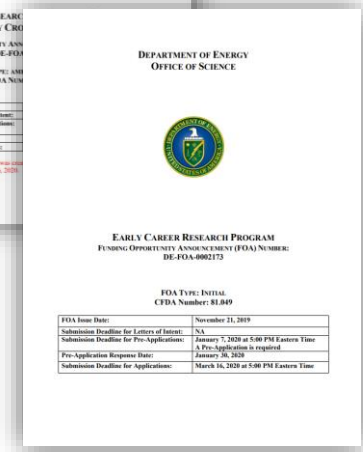


DE-FOA-002173

EARLY CAREER RESEARCH PROGRAM

Proposals due: 3-30-2020

Extension to 3-30-2020



National Laboratory Funding Opportunities FY 2020

2020 DOE Lab Bioimaging Call

Lab Opportunity for proposals to support fundamental research towards exploring new quantum science-enabled bioimaging and/or sensing approaches to achieve an advanced understanding of plant and microbial systems relevant to bioenergy and environmental research.

Proposals Due: **4/13/2020**

Extensions (case-by-case basis)

2020 Lab Opportunity in Secure Biosystems Design Research

Lab Opportunity for new Science Focus Area (SFA)s in Secure Biosystems Design to develop high-throughput, genome-scale design to understand and enhance the stability, resilience, and controlled performance of DOE-relevant plant and microbial systems.

Science Plans Due: 6-30-2020

Update on Programmatic Activities

Completed Reviews/Activities

- ✓ UCLA-DOE Institute Review Oct 29th-31st
- ✓ Bioenergy Research Center (BRC) Annual Progress Reviews
 - CBI Nov 6-7, JBEI Dec 3-4, GLBRC Jan 15-16, CABBI Feb 4-5
- ✓ 2020 Genomic Science Annual PI Meeting Feb 24-26
- ✓ 2020 Bioimaging Science Annual PI Meeting Feb 26-27
- ✓ NCXT review (joint with NIH) Apr 2

Reformatted and/or Canceled Activities due to COVID-1

- Quantum Information Science (QIS) Workshop Mar 12-13 **CANCELED**
- ✓ BNL QPSI *Virtual* Panel Review Mar 17 Completed- **Reformatted to Virtual**
- Early Career Panel review Apr 30 – May 1- **Reformatted to Virtual**
- LAB Bioimaging Science panel review Jun 15 – **Reformatted to Virtual**
- CompBio FOA panel review Jun 16-17 - **Reformatted to Virtual**
- Sustainability FOA panel review Jun 18-19 - **Reformatted to Virtual**
- LAB Secure Biosystems Design panel review Aug 17 - **Reformatted to Virtual**

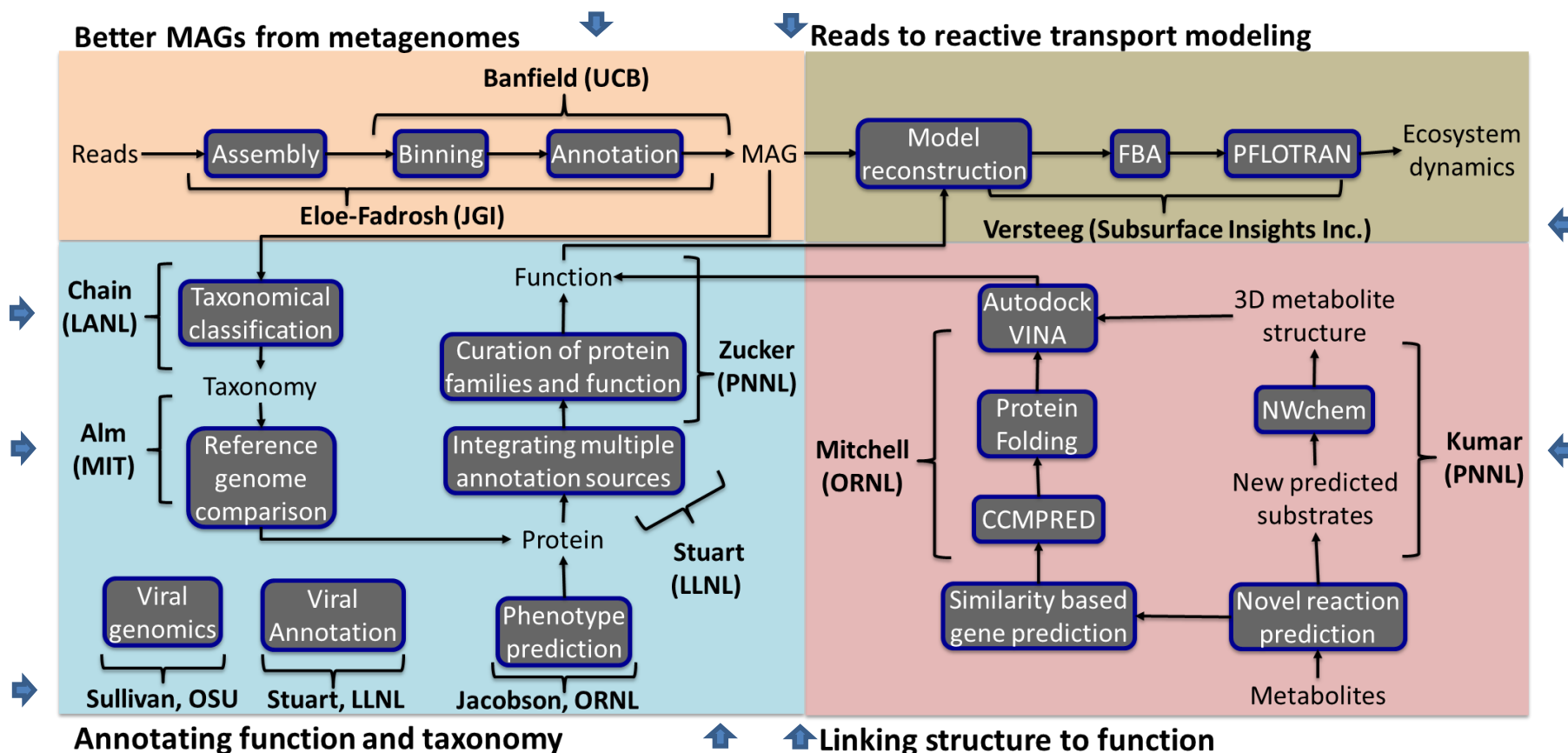
Update on Programmatic Activities Cont'd

Upcoming Reviews/Activities – DATES AND FORMAT PENDING

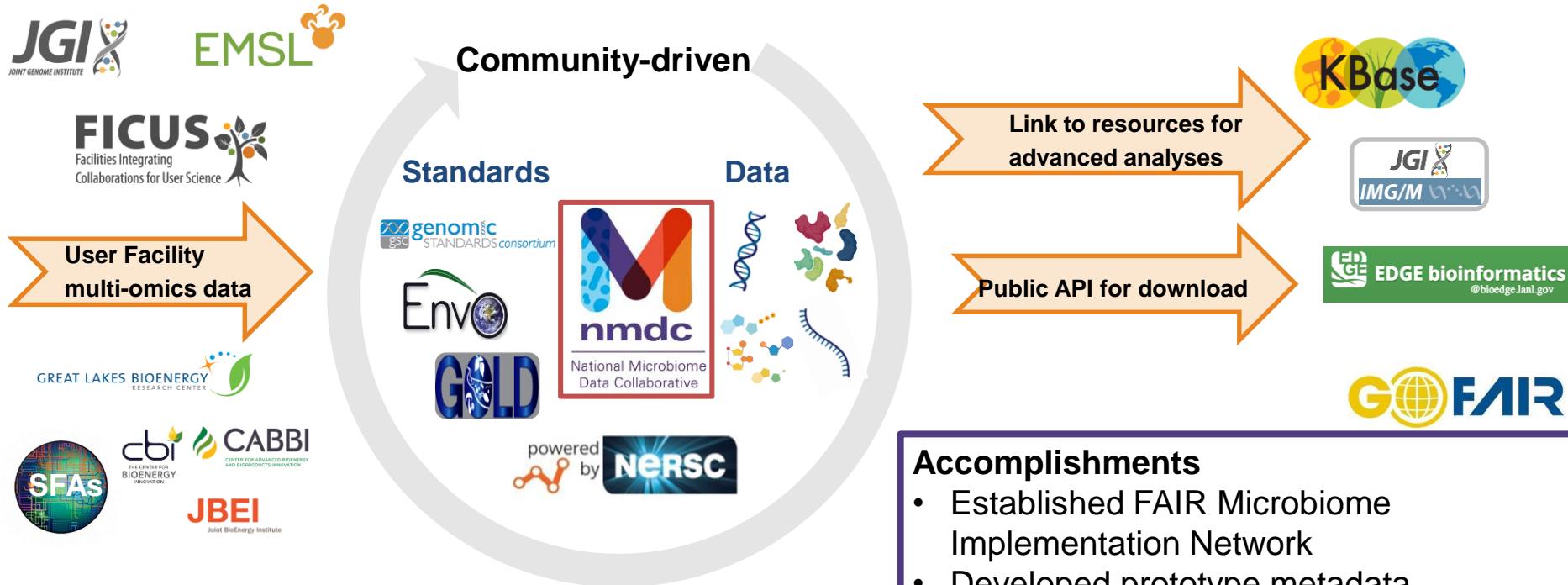
- Committee of Visitors (COV) review Jul 22-23
- National Microbiome Data Collaborative (NMDC) panel review Aug 3
- LBNL ENIGMA Science Focus Area (SFA) panel review Aug 4-5
- LLNL Biofuels Science Focus Area (SFA) panel review Aug 6
- DOE Systems Biology Knowledgebase (KBase) reverse site review Sept 17-18
- Joint Genome Institute (JGI) Triennial review (on site) Dec 1-3

Improving Metagenomic Assembled Genomes (MAGs) for Functional Use

Separately funded efforts to build out KBase capabilities for use as tools with the Lab Science Focus Areas and FOA-funded projects



Piloting an Integrative Data Ecosystem



Metadata

<https://github.com/microbiomedata/nmDC-metadata>

Software

<https://github.com/microbiomedata/WorkflowPlanning>

<https://hub.docker.com/u/microbiomedata>

Accomplishments

- Established FAIR Microbiome Implementation Network
- Developed prototype metadata search interface
- Established NMDC Code of Conduct and Data Use Policy
- Initiated community-centered iterative design process



Cellulose Degradation – a Novel Cellulase

Objective

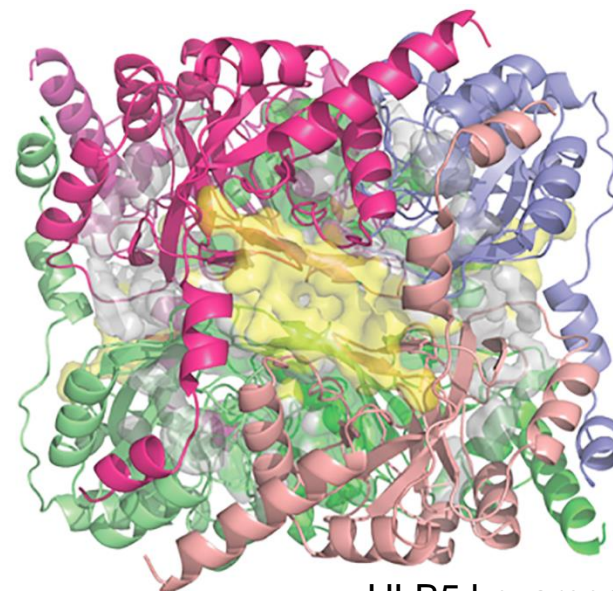
UCDavis researchers identified, synthesized and expressed a putative cellulase gene from microbial dark matter (HLB5) with high endoglucanase activity. Traditional bioinformatics analysis predicted methyltransferase activity for this protein.

Approach

Using x-ray crystallography methodology, SBC team produced and crystallized enzyme, collected data and determined high-resolution structure of HLB5.

Results/Impact

- Structural analysis revealed a novel cellulase with an active site highly similar to CelC endoglucanase from *Clostridium thermocellum*.
- Its structure is different from and doesn't possess conserved regions typical for CAZymes.
- HLB5 has little sequence similarity with known cellulases but possesses high cellulolytic activity against carboxymethylcellulose and pretreated Miscanthus.
- HLB5 is a novel cellulase and can contribute to the overall catalogue of carbohydrate-active genes and proteins currently available.



HLB5 hexamer

NIH Funded project

Chang *et al.* *Protein Science*, 2019

Diversion of Carbon Flux from Sugars to Lipids Improves the Growth of an Arabidopsis Starchless Mutant

Objective

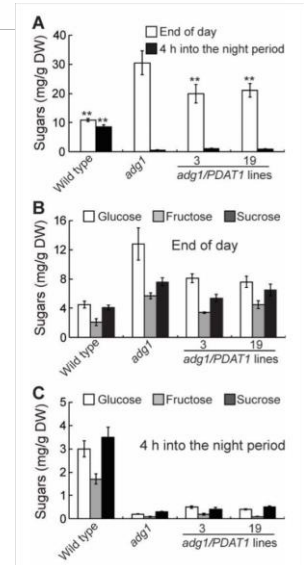
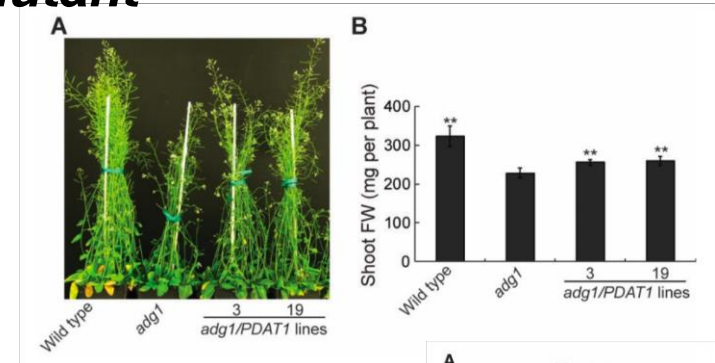
- To test if redirecting carbon from seed carbohydrate to vegetative lipids relieves growth inhibition.

Approach

- To test the role of lipids in plant growth, transgenic plants overexpressing PDAT1 in *adg1* mutants were generated and grown under a 16-h/8-h day/night cycle.

Results/Impacts

- Overexpression of PDAT1 enhances fatty acid and TAG synthesis at the expense of soluble sugars.
- Lipids in the form of TAG can partially replace the function of starch in maintaining energy homeostasis and plant growth in starchless mutants.
- This study improved understanding of how increasing TAG accumulation affects plant growth.



Fan et al. 2019. "Diversion of Carbon Flux from Sugars to Lipids Improves the Growth of an Arabidopsis Starchless Mutant." *Plants*. 8(7): 229. DOI: 10.3390/plants8070229.

This effort jointly supported by BES Physical Biosciences

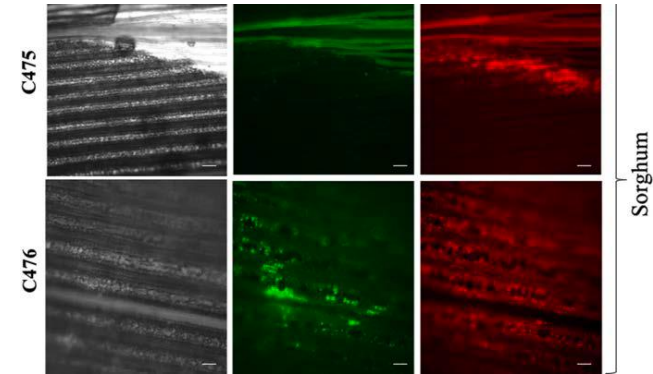
Agrobacterium-mediated transient expression in sorghum leaves for accelerating functional genomics and genome editing studies

Objective

- Develop a transient gene expression system in sorghum to efficiently screen promoter-gene constructs

Approach

- Develop an Agrobacterium-mediated transient gene expression assay with intact sorghum leaves using green fluorescent protein as marker
- Test genome editing capability using sgRNA to GFP protein in sorghum



Testing of sgRNAs for genome editing. Green fluorescence shows editing of frame-shifter GFP (middle panel), while red fluorescence is a reference for successful transformation. C476 effectively causes editing while the negative control C475 does not result in green fluorescence.

Results/Impacts

- In planta Agrobacterium-mediated transient expression of transgenes is achievable in sorghum leaves
- High reproducibility, simplicity, rapidity and feasibility to transform large constructs, which can directly be used for stable transformation
- This method can be used for subcellular localization studies and physiological assays

Sharma et al. (2020) *BMC Research Notes*, doi: 10.1186/s13104-020-04968-9

Identification of critical mutation in a novel regulator of lignin synthesis in poplar

Objective

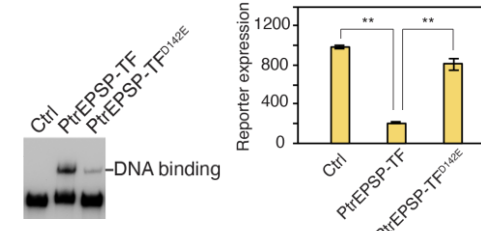
- Identify functional SNPs in *Populus trichocarpa* PtrEPSP-TF responsible for a novel transcriptional repressor regulating phenylpropanoid (PP) biosynthesis.

Approach

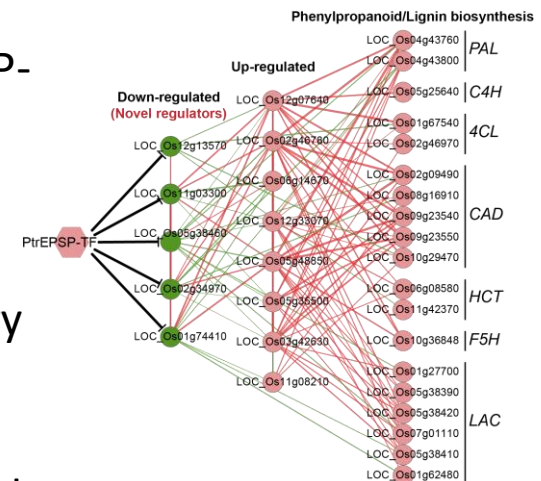
- Used GWAS studies to predict high-impact SNPs in PtrEPSP-TF.
- Characterized transgenic lines heterogeneously expressing PtrEPSP-TF and performed phenotypic and transcriptomic analyses to discover downstream regulators in the PP pathway.

Results/Impacts

- Defined SNPs at the 142nd amino acid of PtrEPSP-TF as the primary determinant of function.
- Uncovered 5 new TF negatively regulating PP biosynthesis.
- Provides molecular targets for precision genome-editing to regulate PP biosynthesis to improve biomass feedstocks characteristics.



SNP-induced substitution of the 142nd amino acid (D142E) dramatically impairs (A) the DNA binding activity and (B) the transcriptional repressor activity of PtrEPSP-TF.



PtrEPSP-TF-mediated transcriptional network. Green edges indicate negative co-expression. Red edges indicate positive co-expression. Black edges indicate experimental validated regulation.

Xie, M., Zhang, J., Singan, V., McGranahan, M., LaFayette, P., Jawdy, S., Engle, N., Doeppke, C., Tschaplinski, T., Davis, M., Lindquist, E., Barry, K., Schmutz, J., Parrott, W., Chen, F., Tuskan, G., Chen, J.G., Muchero, W. (2020). *Plant Direct* 4: 1-13. doi: 10.1002/pld3.178

Nitrate leaching in perennial systems compare favorably to corn crops over time

Objective

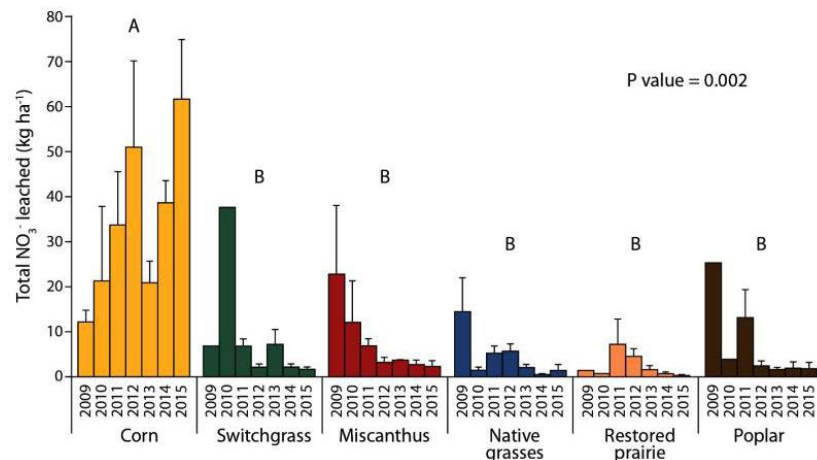
- Compare nitrate (NO₃⁻) leaching of perennial crops to corn over a 7-year period between 2009 and 2016.

Approach

- Soil water was sampled from below the root zone using suction cup samplers.
- Leaching was estimated from nitrate concentrations in soil water and modeled drainage rates.
- Annual summaries represent crop years (harvest to planting) beginning with 2009 harvest

Results/Impacts

- Over 7-years, perennial grasses and poplar trees leached much less nitrate than corn.
- Nitrate leaching from grasses and poplar was comparable with corn for 1–2 years after planting.
- Perennial cropping systems can reduce nitrate pollution in agricultural landscapes.



Total nitrate leached in six cropping systems measured (kg N ha⁻¹ yr⁻¹).

M.Z. Hussain, *et al.* "Nitrate Leaching from Continuous Corn, Perennial Grasses, and Poplar in the US Midwest." *Journal of Environmental Quality* (2019). [DOI: 10.2134/jeq2019.04.0156]

Environmental Microbiome and C Cycling

Metatranscriptomic reconstruction reveals RNA viruses with the potential to shape carbon cycling in soil

Objective

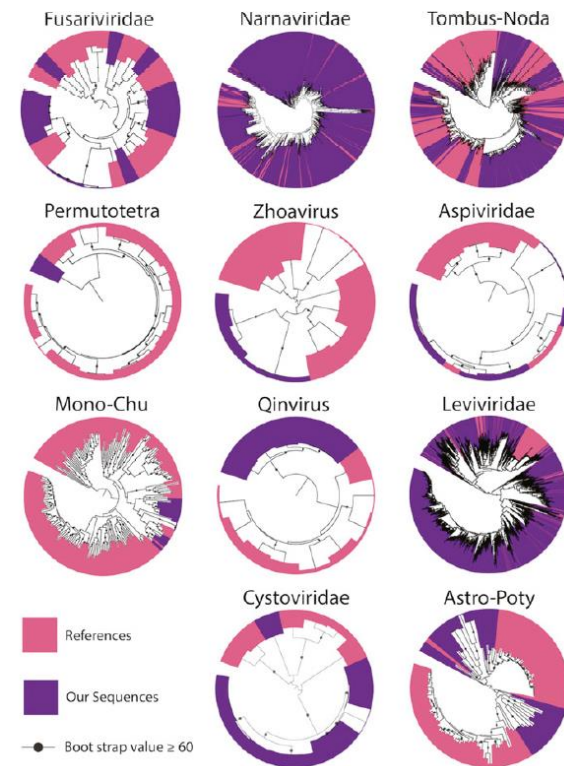
- Determine the impact of RNA viruses on soil carbon cycling

Approach

- Microcosm studies of soil from a California annual grassland field site with and without root litter over a time series (3, 6, 12, and 22 days)
- Identified RNA viral signatures. Analyzed viral and potential host diversity, and transcript abundance

Results/Impacts

- Metatranscriptomes analysis suggested the most common RNA viruses in soil infect fungi (mycoviruses) as well as alpha- and gammaproteobacteria
- 50% of identified viral sequences belonged to the mitoviruses, which replicate in fungal mitochondria
- The presence of root litter, but not roots, influenced viral community composition
- RNA viral communities respond rapidly to resource availability.



PNAS 2019
doi/10.1073/pnas.1908291116

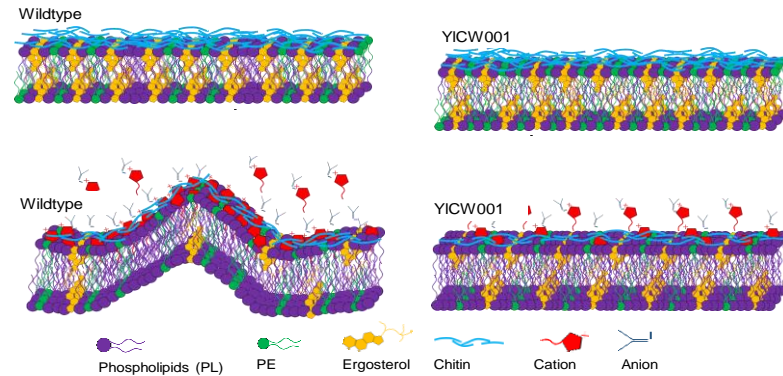
Rearranging membranes improve solvent tolerance in *Yarrowia lipolytica*

Objective

Determine how the oleaginous yeast *Yarrowia lipolytica* grows robustly with enhanced tolerance to high levels of ionic liquids (IL).

Approach

- Generated improved IL-tolerance of *Y. lipolytica* through adaptive laboratory evolution with increasing concentrations of IL.
- Performed characterization and omics analyses to elucidate tolerance mechanisms.



Results/Impacts

- The mechanisms of IL toxicity and superior tolerance in *Y. lipolytica* are not fully understood.
- Adaptive laboratory experiments resulted in strain YICW001 with robust growth in 18% (v/v) IL and tolerance to high concentrations of 7 different ILs lethal to the wildtype strain.
- Micrographs revealed ILs cause membrane disruption, but YICW001 showed little change to cellular morphology.
- Sterols revealed as critical membrane components conferring high IL-tolerance.

Enabling Biomanufacturing Through Multiple Microbial Hosts

Objective

A new technology that allows synthetic biologists to domesticate any bacterial host and, in a single step, successfully and efficiently insert constructs.

Approach

- JGI developed a chassis-independent recombinase-assisted genome engineering (CRAGE) tool that allowed inserts from 10 kilo basepairs (Kbp) up to 48Kbp on nearly 30 gammaproteobacteria species

Results/Impact

- CRAGE saves time as constructs can be inserted in a single step in one day instead of having serial inserts over several days.
- Novel secondary metabolites not normally produced by the hosts were uncovered. Secondary metabolites are the basis for hundreds of invaluable agricultural, industrial, and medical products.
- CRAGE can be adapted to other organisms such as fungi and archaea.

Wang G et al. [CRAGE enables rapid activation of biosynthetic gene clusters in undomesticated bacteria](#). *Nat Microbiol*. 2019 Oct 14. doi: 10.1038/s41564-019-0573-8.

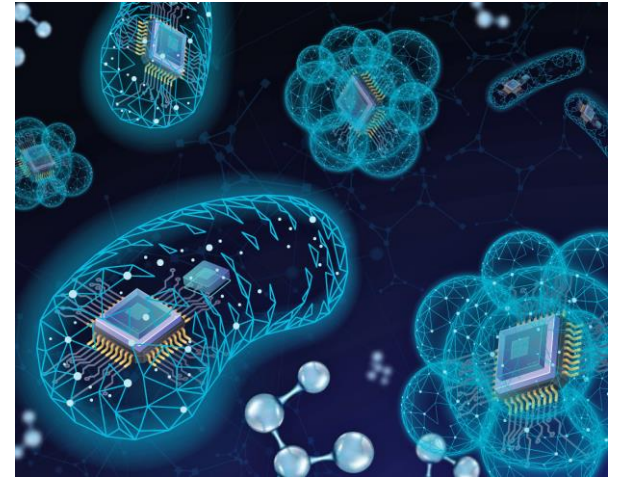


Illustration imagining the molecular machinery inside microbes as technology.
(Wayne Keefe/Berkeley Lab)

Biosystems Design Highlight

A MAGIC (multi-functional genome-wide CRISPR) approach to map genotype to phenotype

Objective

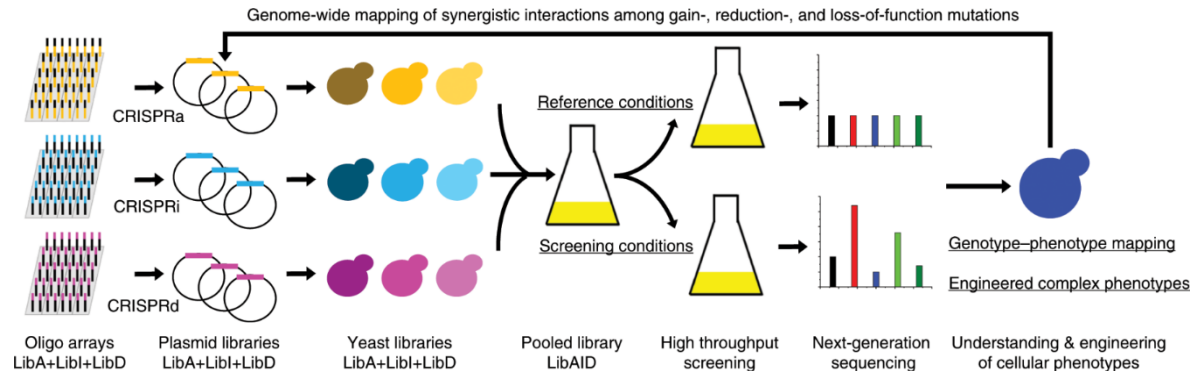
Develop a high throughput system for altering the expression of every gene in the yeast genome to identify the genetic determinants of complex traits

Approach

Combining CRISPR-based activation, interference, and deletion, in a comprehensive yeast mutant library was generated to identify genes responsible for specific phenotypes.

Result/Impacts

- MAGIC was developed to increase, decrease, and eliminate expression of every yeast gene.
- Contains a library of 100,000 guide RNAs redundantly targeting all 6,000 yeast genes.
- Groups of genes responsible for complex traits can be readily identified using this method
- Three mutations identified that confer tolerance to furfural.
- MAGIC was also used to generate strains with increased activity of a heterologous cellulase expressed in yeast.



Lian *et al.*, *Nat Commun* 10, 5794 (2019)

Plant cell-surface GIPC sphingolipids sense salt to trigger Ca^{2+} influx

Objective

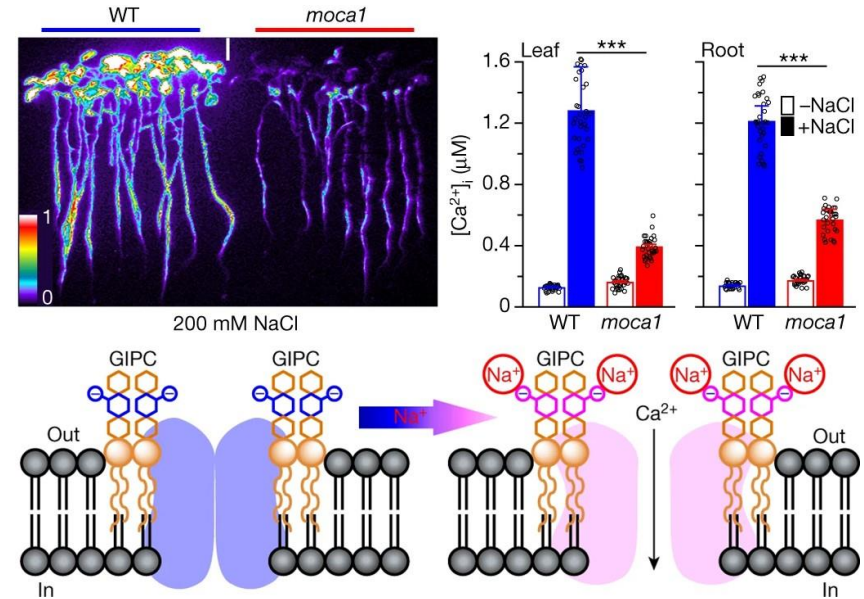
How do plants recognize salt in their surroundings and trigger Ca^{2+} signals that lead to salt tolerance.

Approach

Use aequorin bioluminescence-based Ca^{2+} imaging to screen for *Arabidopsis* mutants impaired in increases in Ca^{2+} induced specifically by ionic but not osmotic stresses.

Results/Impact

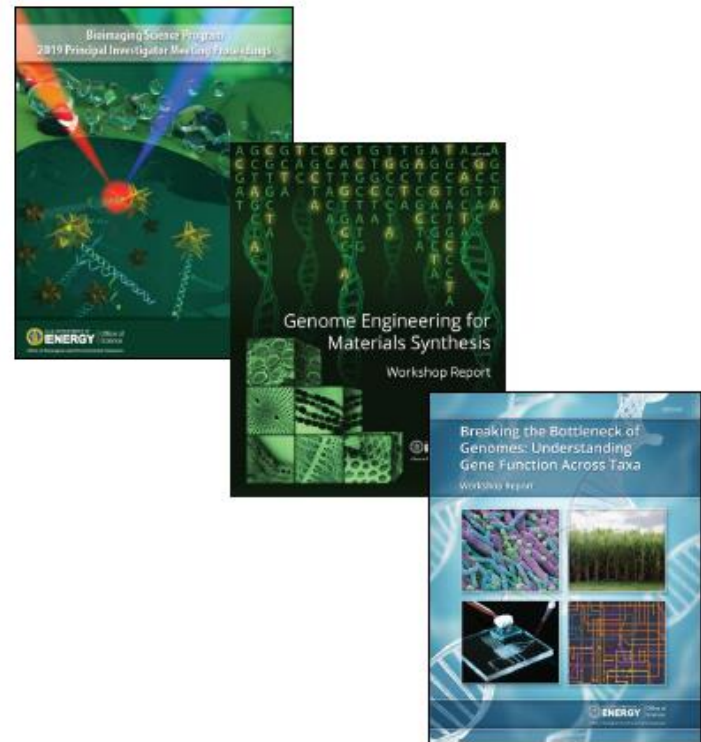
- Isolated *Arabidopsis* mutant with an abnormally low Ca^{2+} -signaling response to high salt.
- Identified MOCA1 as a glucuronosyltransferase for glycosyl inositol phosphorylceramide (GIPC) sphingolipids in the plasma membrane that can bind Na^+ ions and trigger Ca^{2+} signals that lead to salt tolerance.
- This study reveals novel insights into plant salt sensing mechanism and demonstrates the importance of GIPCs for the regulation of signaling processes at the plasma membrane.



Jiang, Z. et al. 2019. "Plant cell-surface GIPC sphingolipids sense salt to trigger Ca^{2+} influx" *Nature* 572, 341-346, DOI: 10.1038/s41586-019-1449-

April 2020 BERAC Meeting

Thank you



<http://genomicscience.energy.gov>