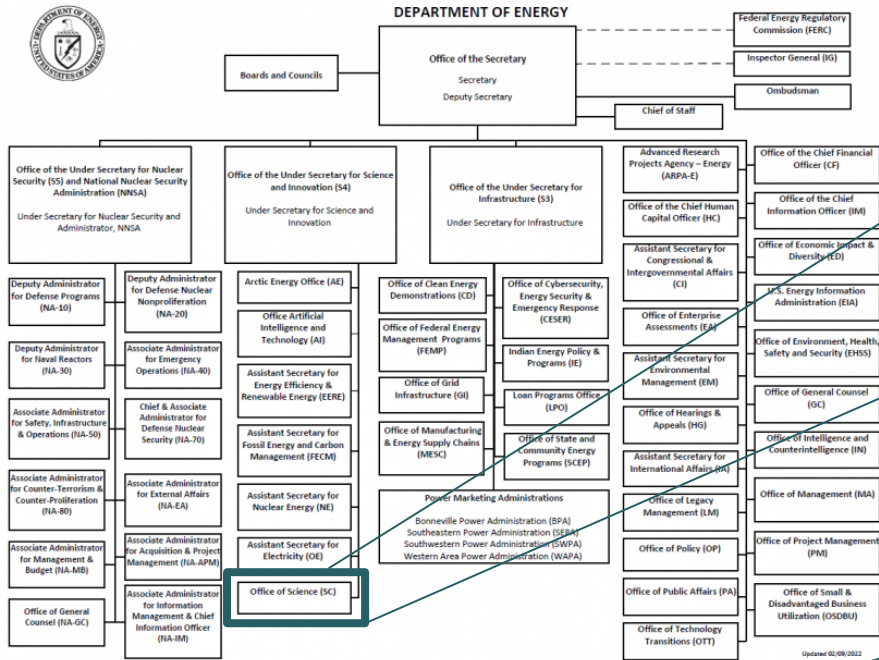


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# Microbiome Research for Carbon Cycling and Sustainable Bioenergy Feedstocks in BER's Genomic Science Program

***Boris Wawrik***

U.S. Department of Energy (DOE), Office of Science (SC)  
Biological and Environmental Research (BER)  
Biological Systems Science Division (BSSD)



Integrate observations and experimental capabilities for predictive systems-level understanding, from microbes and plants to ecosystems and the earth system

**Biological Systems Science Division**  
 Division Director: Todd Anderson

- Bioenergy Research Centers
- Genomic Science Program
- Biomolecular Characterization
- Imaging Science
- Structural Biology
- Joint Genome Institute

**Earth and Environmental System Science**  
 Division Director: Gary Geernaert

- Atmospheric System Research
- Environmental System Science
- Climate & Earth System Modeling
- Facilities & Infrastructure
- Environmental Molec. Sciences Lab
- ARM Climate Research Facility



# Biological Systems Science Division (BSSD)

**MISSION:** to provide the necessary fundamental science to understand, predict, manipulate, and design biological processes that underpin innovations for bioenergy and bioproduct production and enhance understanding of natural, environmental processes relevant to DOE.

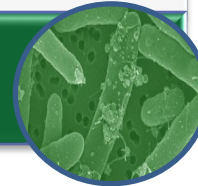
- Bioenergy Research Centers
- Sustainable Bioenergy
- Feedstock Development
- Plant Genomics
- Deconstruction
- Microbial Conversion

Bioenergy Research



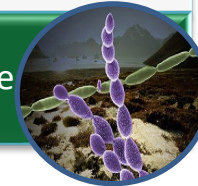
- Synthetic Biology
- Innovative ways for bioproduction of fuels, chemicals, and biomaterials
- Modeling and HTP DBTL
- Secure Bidesign

Biosystems Design



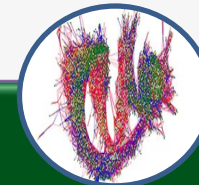
- Microbial Systems Biology
- Natural and Synthetic Microbial Consortia
- Techniques and Technology

Microbiome Science



- Computational Biology
- Biomolecular Characterization and Imaging Science
- DOE User Facilities (JGI, EMSL, Light & Neutron Sources)

Enabling Capabilities

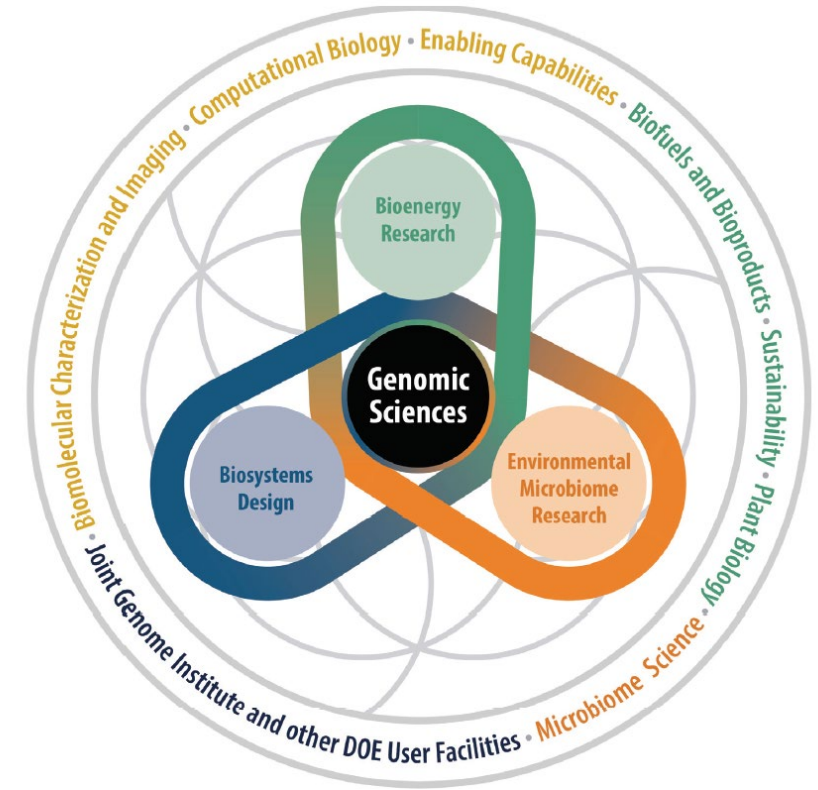


U.S. DEPARTMENT OF  
**ENERGY**

Office of  
Science

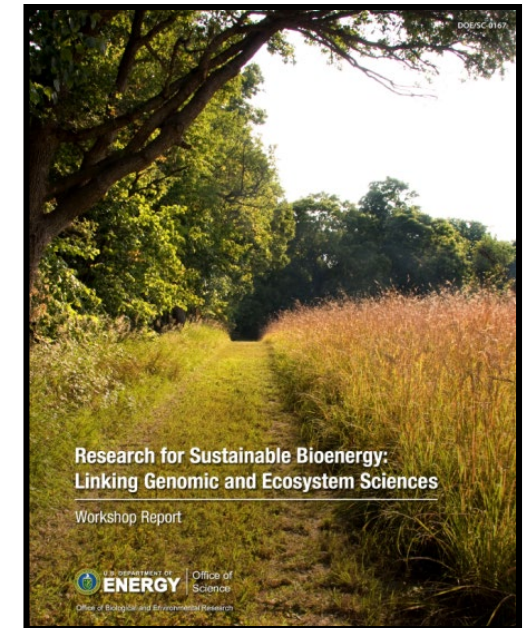
# The Genomic Science Program (GSP)

- What information is **encoded in a genome sequence**, and how can genomes be interpreted to explain the functional characteristics of cells, organisms, and whole biological systems?
- How do interactions among **cells regulate the functional behavior** of living systems, **and how can these interactions be understood dynamically and predictively**?
- **How do plants, microbiomes, and communities of organisms adapt and respond to environmental conditions** (e.g., temperature, water, nutrient availability, and ecological interactions), and how can their behavior be manipulated toward desired outcomes?
- What **organizing principles** need to be **understood to facilitate the design and engineering** of new biological systems for beneficial purposes?



# Sustainable Biomass Production - Research to Advance Bioenergy Agriculture

- Integrated lab and field experiments (field work required)
- **Enhance biomass productivity**
- The **impacts** of bioenergy cropping systems **on the local ecosystem**
- Impact of **changing environmental conditions**
- Molecular and physiological mechanisms that control
  - **bioenergy crop vigor**
  - **resource use efficiency**
  - **resilience/adaptability to abiotic stress**
- Role of **microbial communities** in plant-soil environment
- **Microbiome role in plant performance**
  - What role does the microbiome play in plant performance ?
  - What are the mechanisms of plant-microbe interactions ?
  - How can microbial traits be leveraged to enhance sustainability?

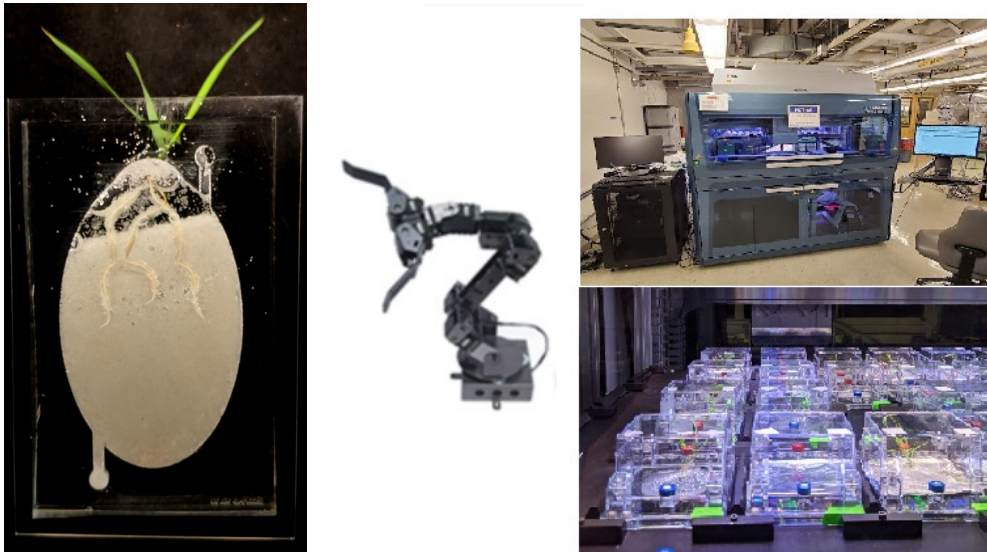




# Example - Technology Integration to Standardize Microbiome Experiments

## EcoFABs

- Controlled model ecosystems in which microbes and host responses can be monitored
- High replicability
- Allows automation
- Can integrate imaging systems approaches



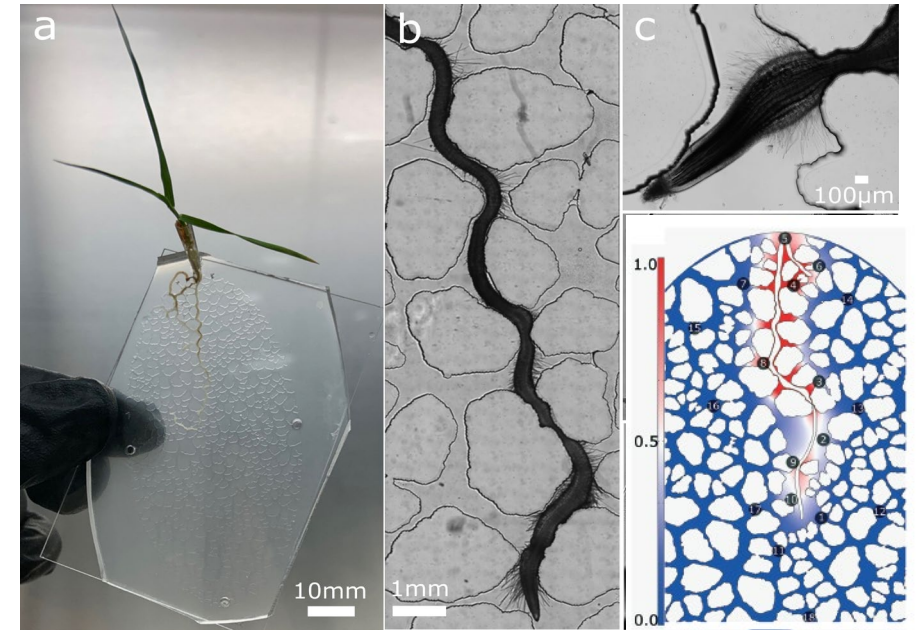
An EcoFab device containing artificial soil and a plant seedling. HTP sampling and detection can be achieved with the EcoBOT.

<https://eco-fab.org/>



## Rhizosphere-on-a-chip

- Tractable approach for studying carbon hotspot formation and plant-microbe interactions in soils.
- Create a synthetic soil habitat that enables dynamic imaging and spatial chemical sampling of plant roots.

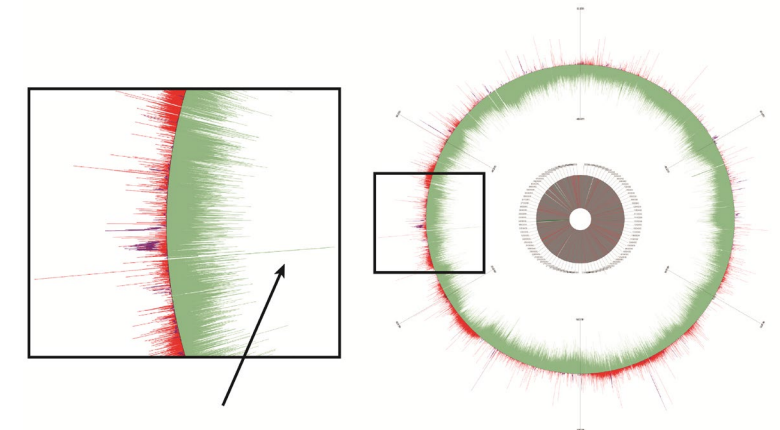
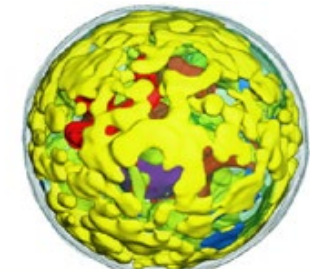
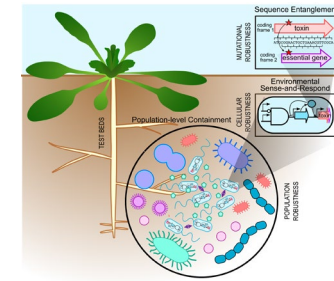


A *Brachypodium* seedling growth in the rhizosphere-on-a-chip. Hotspots of exuded amino acids are detected within the rhizosphere.



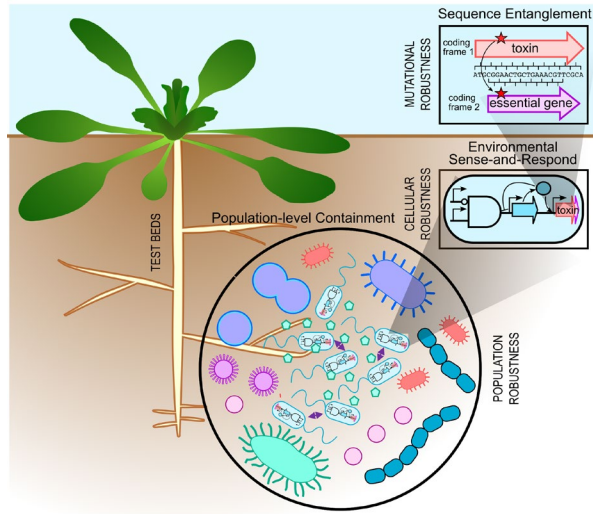
# Biosystems Design – Systems Biology and Genome Engineering Basic Research

- Design and **engineer new biological systems**
- Innovative bioproduction of **fuels, chemicals, and biomaterials**
- Engineer **microbes, plants, and microbial consortia**
- **Genome-scale engineering**
- Develop novel ***in vivo* and cell-free engineering tools**
- Create new biological functions
- Development of **new platform organisms** for genome engineering
- High-throughput approaches for **screening and testing**
- Upcycling of synthetic polymers
- **Secure bio-design**



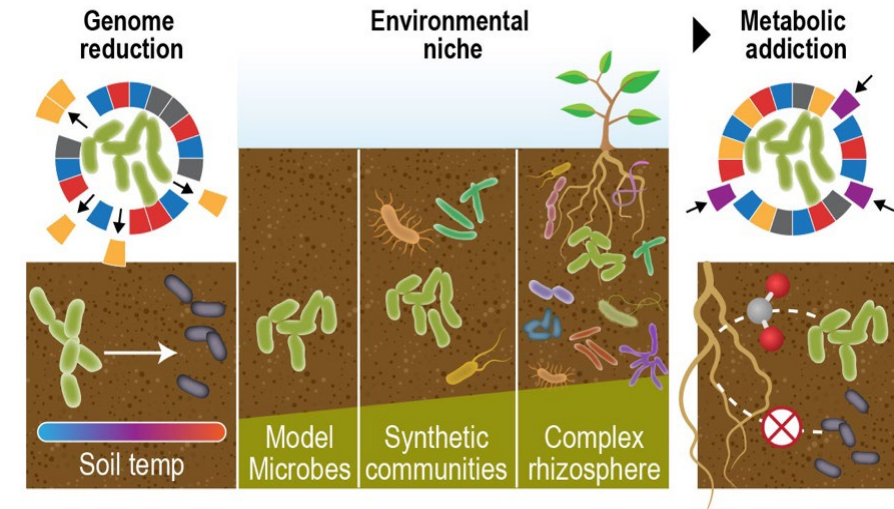
# Examples - Secure Biosystems Design Projects

## Secure and Robust Biosystems Design for Environmental Microorganisms (LLNL)



- reduce the risk of unintended ecological consequences from environmentally deployed genetically engineered microorganisms (GEMs)
- design multilayered containment strategies for GEMs in the rhizosphere
- advance synthetic gene entanglement strategy for containment

## Persistence Control of Engineered Functions in Complex Soil Microbiomes (PNNL)

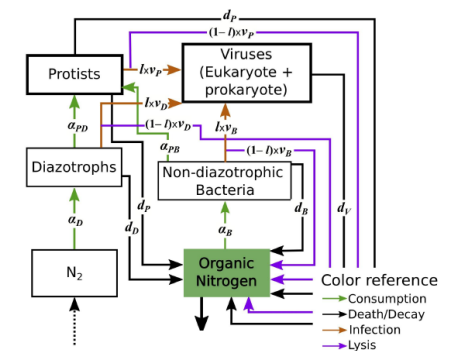
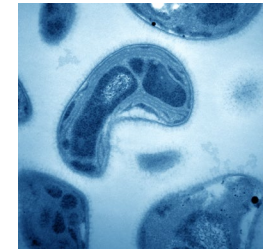
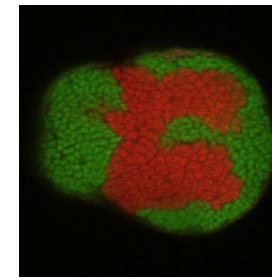
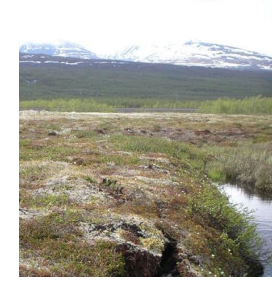


- design the environmental niche of bacteria through genome reduction
- determine the genetic, metabolic, and spatial factors that control microbial persistence in the rhizosphere
- develop genetic tools for phylogenetically diverse rhizosphere bacteria
- Design metabolic addiction strategies for GEMs



# Environmental Microbiome Research

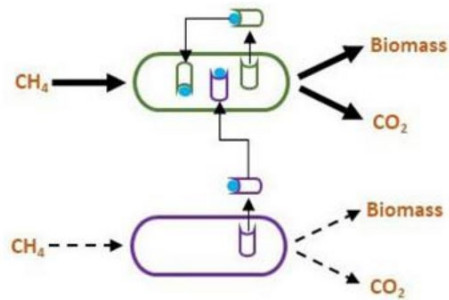
- Microbial activity relevant to **biogeochemical processes**
- Systems-level research on **nutrient and carbon cycling**
- **Mechanistic** underpinnings of **microbial activities**
- Interactions of **viruses, bacteria, archaea, plant, fungi, and protists**
- ‘omics-based high-resolution, high-throughput techniques and technologies
- Integrate microbial processes across scale via **computational and modeling approaches**
- Develop a predictive understanding of microbial systems behavior in terrestrial **soil ecosystems** -- including the cycling, release, and storage of C in soils



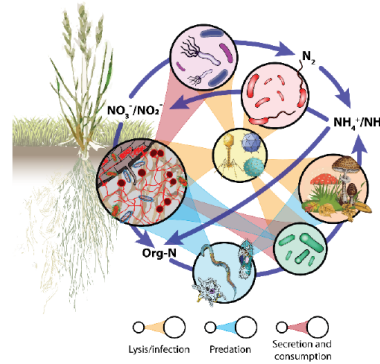
# Soil C-cycling Projects in the Environmental Microbiome Portfolio



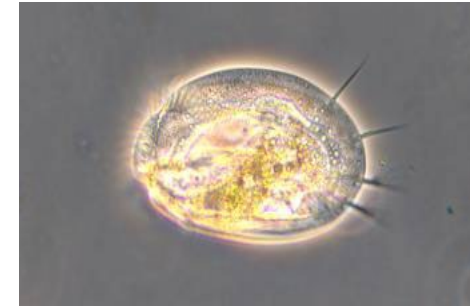
Microbial degradation of **pyrogenic organic matter**



Impact of elemental cycles e.g. **P** or **Cu** on **C and N cycles**



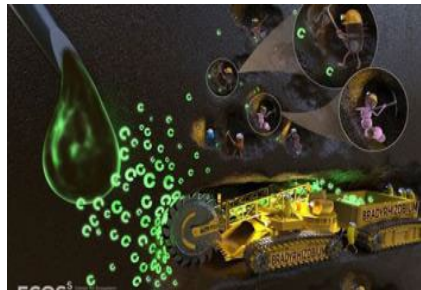
**Interkingdom interactions** in soil **C cycling**



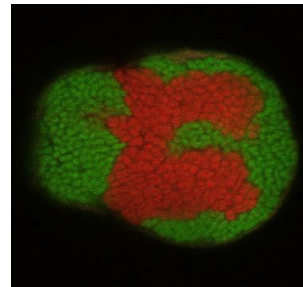
The role of **protists** in organic matter turnover



The role of **viruses** in soil C cycling



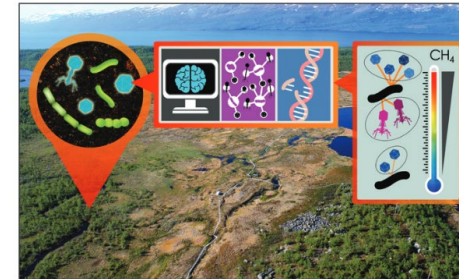
Microbial traits and models of **behavior at the macro scale**



Tools to study anaerobic **methane metabolism**



**Microbial adaption and evolution** to **drought** in soils



Microbial C cycling in thawing **permafrost**

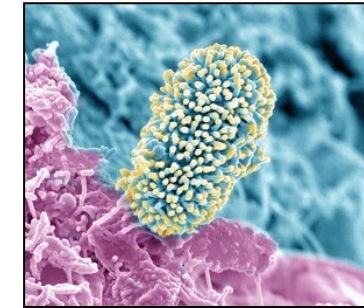


**Metabolic exchange** in soils, wetlands, and artificial ecosystems

# The Joint Genome Institute (JGI)

- DNA **sequencing** of fungal, algal, bacterial, archaeal, viral and plant genomes; community shotgun DNA/RNA; 16S;
- Whole genome DNA **methylation** analysis
- **Transcriptomics**; non-coding RNA (both small and long ncRNA) characterization;
- Fluorescence activated cell sorting; **single-cell genomics**
- **DNA/gene synthesis**; assembly of biosynthetic pathways in heterologous hosts
- CRISPR-based **gRNA library construction** and QC.
- Mass spectrometry-based **metabolomics**
- **Analysis** pipelines for the datasets above
- **Technology Development**

Support projects through **Community Sequencing Program (CSP)** Call.  
Collaborative funding with EMSL via the **JGI-EMSL FICUS** (Facilities Integrating Collaborations for User Science) program.



Root associated  
microbe



*Eucalyptus grandis*



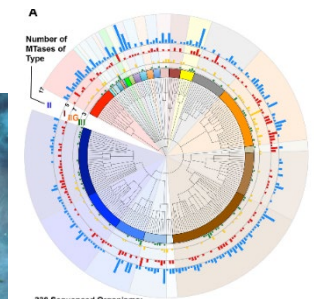
*A. muscaria*



*Setaria viridis*



Updated: Tree of Life



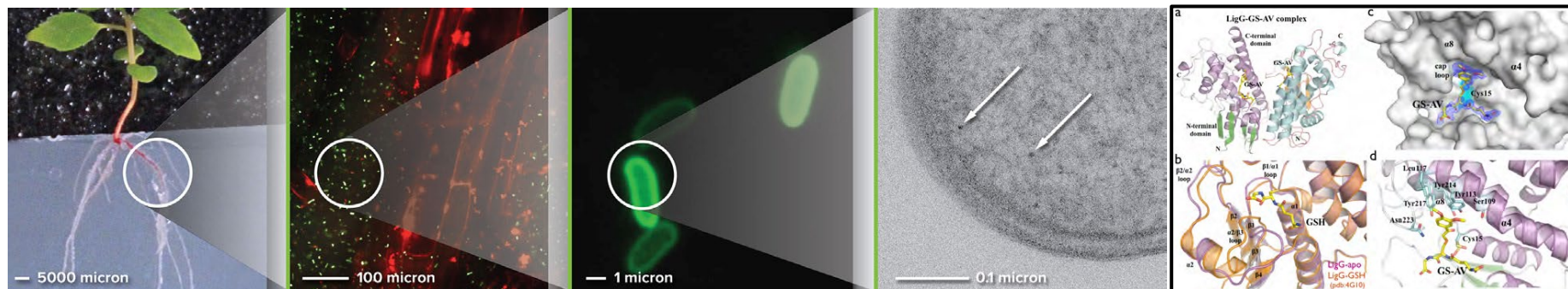
Epigenomics



# Bioimaging and Characterization

## Visualization across scales of observation to biomolecules

- Develop *in situ*, dynamic, and nondestructive multifunctional imaging
- Create **enabling visualization and characterization capabilities**
- Combine biomolecular structural characterization with genomics information and bioinformatics **to infer function and improve genome annotation** or design new functions.
- **Visualize expressed biomolecules** within living plant or microbial cells or within microbial communities.
- **Quantum imaging**





# Computational Biology

*Goal is to support the development of computational and instrumental platforms to enable broader integration and analysis of large-scale complex data within BER's multidisciplinary research efforts.*

- Assemble capabilities for the processing of **large**, complex, and **heterogeneous systems biology data**.
- Create the next generation data systems and algorithms that connect observations across scales of **molecular, structural, genomic**, and other omics data with cellular and multicellular processes.
- Develop explainable **artificial intelligence** (XAI) algorithms to identify relationships among different parts of genomes
- Create techniques to process and **integrate imaging and structural biology data** with simulation and other biological measurements.
- **Advanced simulation capabilities** to model key processes occurring within or among cells building towards **whole-cell simulation**.
- Assemble an **integrated systems biology virtual laboratory** to accelerate *in silico* ideation and collaboration within the research community.



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# QUESTIONS