



Office of Science

Project Initiation at the JGI

Project Management Office



Project Management Office (PMO) Team



Tootie Tatum, Group Lead



Kerrie Barry, Plant and Fungal Genomics Programs



Natasha Brown, Supply Chain Manager



Tijana Glavina del Rio, Metagenomics Program



Nancy Hammon, Workflow Planning Manager



Miranda Harmon-Smith, DNA Synthesis and Single-Cell Genomics



Vivian Ng, Project Manager



Christa Pennacchio, Special Projects



Nicole Shapiro, Microbial Genomics Program

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WHAT'S HAPPENING NOW AT JGI



Latest JGI News

SUBSCRIBE

MARCH 4, 2015

Characterizing Permafrost Microbes in a **Changing Climate**

ALL NEWS RELEASES

Announcements

Letters of Intent Now Accepted for JGI-EMSL Collaborative Science Initiative - April 6 deadline

The Community Science Program (CSP) is now accepting Letters of Intent for large-scale sequence-based genomic science projects, deadline April 16, 2015.

JGI hosting 17th Genomic Standards Consortium meeting May 4-6 and Genomes to Secondary Metabolites workshop May 7-8, 2015







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Product Offerings

Scientific Program	Product	Brief Description	Deliverables	FY15 target cycle time (median), days	FY15 target cycle time (75th %), days
Fungal	Minimal Draft	Low coverage whole genome shotgun sequencing for evaluation. May turn into a standard draft or improved standard draft.	Assembly. Annotation optional (JGI portal); raw data submitted to SRA	250	400
Fungal	Resequencing	SNP and short indel calls, rearrangement detection, population analysis.	Text file of SNPs (incl location in genome, coding/vs non, syn vs non-syn aa change etc) and structural rearrangements, alignment files, tracks for upload to genome browser and fastq files; raw data submitted to SRA	140	200
Fungal	Standard Draft	Whole genome shotgun sequencing. Exact scope items and quality of finished product depend on genome. Selected genomes will be improved based on feasibility and scientific merit.	Assembly, annotation (JGI Portal + Genbank); raw data submitted to SRA	250	400

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MyJGI Home

Proposal:

Log in

Submit a sequencing or synthesis proposal to the JGI:

The Work Initiation Process (WIP) application provides a web-based interface that can be used to request sequencing or synthesis from the JGI (view current product list). The current release supports submissions of all proposal types below.

JGI-EMSL Collaborative Science Initiative:

This user program is open to everyone during the annual call. The JGI-EMSL 2016 call is now open and Letters of Intent are being accepted until April 6, 2015 at EMSL. About JGI-EMSL joint call

Community Science Program (CSP) annual call:

This user program is open to everyone during the annual call. The CSP 2016 call for Letters of Intent is now open until April 16, 2015 23:59 PDT. About CSP

Community Science Program (CSP) small-scale microbial and metagenome:

This program is open to anyone. Small-scale proposals for microbial and metagenome projects may be submitted at any time as brief white papers and will be reviewed every six months. Proposals may include microbial isolates, RNA, resequer days prior to the review. See <u>proposal schedule</u> for specific dates. <u>About CSP</u>

Community Science Program (CSP) synthetic biology:

This program is open to anyone. Proposals for synthesis projects may be submitted at any time as white papers and will be reviewed every six months. Deadline for submission is 60 days prior to the review. See proposal schedule for specific days.

Bioenergy Research Centers (BRCs):

This program is open to PIs associated with DOE Bioenergy Centers only. Proposals may be submitted at any time. About BRCs

All users must create a "JGI Single Sign-On Account"



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Start a new proposal...

CSP 2016 Annual Letter of Intent

Begin

CSP small-scale microbial and metagenome

You can see an example of such proposal here.

Begin

CSP synthetic biology

Begin

Bioenergy Research Center

New proposal: select a focus area \$ Begin

All other proposal types (Director's Science/R&D/WFO)

New proposal: select a focus area Begin





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CSP2016 call

New CSP 2016 Letter of Intent

Fitle * 255 characters	
_	sion relevance appropriate to your proposal
bioenergy	
biogeochemistry	
bioremediation	
carbon cycling	
 phylogenetic diversity 	
Relevant categories	
Please select one or more categories to	nat are relevant to your proposal.
 Extreme Environments 	
 Chromatin analysis 	
 Plant flagship genomics 	
 Plant microbiomes 	
 Algal genomics 	
 Microbial function 	
Metagenomics	
Fungal genomics	
Synthesis	
Proposal summary	
Description * 4000 characters	
Please briefly describe sequencing or s	synthesis needs for your proposal. Include estimated genome size for each whole genome shot
organism, or estimated total resource n	eeds for other types of projects. Please refer to sequencing and synthesis limits at
http://jgi.doe.gov/collaborate-with-jgi/co	mmunity-science-program/how-to-propose-a-csp-project/csp-annual-call/.
Justification * 4000 characters	
Please briefly describe the scientific rat	ionale for performing this work.



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The DOE JGI Project Management Office (PMO) carries out a number of activities to facilitate world-class science. DOE JGI Project Managers collaborate with users to develop project requirements, define the scope of genomic work and outline roles and responsibilities resulting in a formal Statement of Work (SOW).

All work performed at the DOE JGI is initiated via the Work Initiation Process (WIP) interface. To submit a proposal, users must first log in (Note: new users of the DOE JGI will need to register and create a login at https://signon.jgi.doe.gov).

Once users are notified that their proposal has been accepted, the PMO will assist with initiation of work through:

- Coordinating execution of <u>Institutional User Agreements</u>
- Coordinating with DOE JGI technical experts
- Project design with Principal Investigator
- Providing a General Orientation to DOE JGI systems to Investigators
- Preparation of formal Statement of Work (project roadmap)



Statement of Work for JGI DNA Sequencing Projects

General Information

Title:

Functional genomics of moss-cyanobacteria interactions in boreal forest ecosystems

Abstract

We propose coupled genomics, transcriptomics, and proteomics to characterize the feather moss-cyanobacteria association that covers the understory in boreal forests worldwide and serves as the main form of biological N input to the mature boreal forests. The two predominant and widespread Pleurocarpous feather mosses, i.e., Pleurozium schreberi and Hylocomium splendens, support a wide diversity of filamentous cyanobacteria dominated by the genera Nostoc and Stigonema. Single gene based surveys suggest host specificity and high genetic diversity in this community of filamentous cyanobacteria. Genome sequences of the associated cyanobacteria and the moss host are not available, although genome sequences exist for the distantly-related association-competent cyanobacterium Nostoc punctiforme ATCC 29133/PCC 73102, originally isolated from the cycad Macrozamia.

Our approach targets all levels of the central dogma, as well as organismal and ecosystem scales. Genome sequencing of cyanobacterial isolates collected in natural boreal ecosystems will provide an understanding of the functional potential and diversity of this crucial partner of the associations and provide fundamental knowledge on how the two partners communicate prior to and during colonization. Our first set of genome sequencing samples will consist of ten cyanobacteria associated with feather mosses that have been isolated in pure culture by Co-PI Rasmussen. Our second set of genome sequencing samples will be from cyanobacterial flaments unable to be cultivated in isolation (i.e. Stigonema). The filaments will be removed from the moss by micro-manipulation and genomic DNA will be amplified. For both sets of cyanobacterial genome samples, we assume a genome size of approximately 8-10 Mb for each isolate or environmental colony, based on the genome size of the related N. punctiforme. This estimate includes plasmids that may be important repositories of genetic diversity for these species.

Signaling pathways involved in forming successful associations will be studied using transcriptomics and proteomics of both partners. We will use an experimental setup developed by Co-PI Rasmussen that allows for communication between the partners without colonization. This novel setup allows the initial signaling phase to be differentiated from the subsequent establishment phase of the interaction. P. schreberi will be used as the moss partner and three different cyanobacteria strains will be used: 1) a Nostoc sp. isolate from the moss P. schreberi as a successful interaction, 2) a free-living Nostoc sp. that does not form associations, and 3) the model cyanobacterium N. punctiforme ATCC 29133 because of its well-established genetic system. Samples will be collected for each cyanobacteria and the P. schreberi moss partner under three conditions: 1) separated (in culture/growing separately), 2) with chemical but not physical contact, and 3) during/after colonization. The use of these strains will enable differential transcriptional patterns to be deduced and will identify candidate cyanobacterial genes to test by genetic manipulation. If time allows, altered physical conditions, such as high CO2 and high temperature, will be tested to understand how changing climate may affect these critical ecosystem interactions. The large transcriptomic data set will facilitate global and targeted proteomic analysis at various states of the interaction.

Finally, the metatranscriptome and metaproteome of several distinct natural moss-cyanobacterial communities will be examined. These will be the same communities from which samples used in this proposal have been collected (see Sample Preparation) and will allow for comparisons of gene expression patterns with the laboratory-based association studies. It will also permit correlation of transcriptional profiles from other organisms in the community leading to the formation of successful associations.

Scope of Work:

ISOALTES TOTAL: 6 organisms Improved drafts. PacBio – 10kb library.

SAGs TOTAL: 8

Send JGI the MDA product

Will be a mixed culture (pools of cells) 10-20 cells per MDA reaction

TRANSCRIPTOME

TOTAL: 30

3 cyanobacteria strains, each with 3 conditions, and 3 replicates (=27 samples), plus 3 moss only samples. For the cyanobacterial only samples (18), rRNA depletion will be used. For the samples that have both cyanobacteria and moss together (9), these will get both rRNA and poly-A library prep methods to allow both the cyanobacterial and moss samples to be represented in the transcriptome. For the three moss-only samples (3), these will get only poly-A library prep.

JGI will pilot 2 transcriptomes for feasibility: a moss-only transcriptome and a moss-cyanobacteria transcriptome.



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Please follow the steps outlined below to submit samples for sequencing or synthesis to the JGI. If you have any questions, please contact your Project Manager or pmo_igi_project_management@lists.igipsf.org.

DNA Sequence (Synthetic Biology) Submission

DNA sequence files for all constructs should be emailed to Miranda Harmon-Smith, Synthetic Biology Project Manager.

DNA and RNA Sample Submission

1) Review JGI sample QC requirements and protocols

Obtaining DNA & RNA of suitable quantity and quality has been the rate-limiting step for many projects at the DOE JGI. The quality of the starting material is one of the greatest predictors of a successful sequencing project. It is imperative that users utilize the Qubit system for assessment of DNA or RNA mass prior to shipment to the DOE JGI. These documents (posted with permission) demonstrate the importance of utilizing this platform for nucleic acid quantification in our Illumina and PacBio sequencing workflows and provide guidance as to how to perform sample QC prior to shipment:

- Importance of Sample QC
- Sample Quality and Contamination
- Genomic DNA Sample QC Protocol
- Total RNA Sample QC Protocol
- iTag Sample Amplification QC v1.1 before submitting samples for iTag sequencing, you must first confirm the quality of material through amplification. We strongly suggest that you use this protocol to confirm the ability of all samples to be used for library construction and sequencing of 16s, 18s or ITS.



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Obtain Approval for Shipping

PLEASE NOTE: The JGI will not accept any materials classified above NIH Biosafety Level 2. Before approval to ship is granted, you must have completed submission of all project metadata for all project materials.

Review Shipping Instructions

After receiving approval, please refer to the emailed shipping instructions for shipping your materials to the DOE JGI. The Shipping Checklist below will assist you in verifying that all required information is included. Please note that shipment of materials to the JGI without prior approval or complete documentation may result in destruction of the shipment. Please retain your carrier's tracking information.

- JGI Shipping Checklist
- International Shipments (For shipping packages internationally (including Canada!) please use these instructions.)
- TCSA Form for International Shipments

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Protocols provided by JGI and the JGI user community.

JGI sample QC protocols:

- Genomic DNA sample QC protocol
- Total RNA sample QC protocol
- iTag Sample Amplification QC protocol

DNA preparation protocols:

- JGI Bacterial DNA isolation CTAB-2012
- Isolation_of_Genomic_DNA_from_Phytophthora
- Isolation_of_Melampsora_Nuclear_DNA
- DNA_Isolation_of_Plant Nuclear_DNA
- Populus nuclear DNA purification with Qiagen
- DNA_extraction_from_Activated_Sludge
- Cloning of High Molecular Weight eDNA (soil)
- RNase_I_DNA_Clean_up_protocol
- eDNA _purification (removal of humics)
- High Molecular Weight DNA Extraction from Soil
- http://1000.fungalgenomes.org/home/protocols/

RNA preparation protocols:

DNase Treatment of Total RNA



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