

## Determination of the Roles of Pyrophilous Microbes in the Breakdown and Sequestration of Pyrolyzed Forms of SOM

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### Project Goals:

The frequency of large, high severity wild fires is increasing in the western US and in regions around the world due to long-term fire suppression strategies and climate change [1]. These fires have direct, negative effects on soil carbon stocks through combustion, but they have indirect and potentially positive effects on soil carbon stocks through the production of pyrogenic organic matter (PyOM) that has a long residence time and constitutes a major pool of C in fire-prone ecosystems [2-4]. Soil microbes are likely to be involved with the degradation of all of these compounds, yet little is currently known about the organisms or metabolic processes involved.

**We are dissecting the effects of microbes on post-fire soil carbon dynamics by using a systems biology approach that couples small experimental “pyrococosms”, highly controlled production of <sup>13</sup>C-labeled pyrolyzed substrates, genomics, transcriptomics, stable isotope techniques, and mass spectrometry to address the following objectives:**

*Objectives:*

1. Develop improved genomic and other -omic resources for the dominant microbes of fire-affected soils
2. Determine the temporal response of soil microbes to fire and to PyOM additions
3. Characterize the temporal patterns of degradation of different sub-fractions of PyOM

*Objective 1.* An important component of this research is to develop and improve the genomic, proteomic, and metabolomic resources available for studying the microorganisms that dominate fire-affected soils. The fungal community in a model fire-affected soil has been well characterized, and fungal isolates from this community have been isolated and sequenced by the project team (see talk Monday afternoon). Similarly, early attempts at cultivating members of the bacterial community in these soils has produced >50 bacterial isolates that are able to grow with highly-aromatic PyOM as their primary carbon source. The majority of these isolates have been identified by 16S rRNA genes as members of the *Actinobacteria* phylum with the genera *Streptomyces* and *Pseudonocardia* being most prominent. Isolates are being evaluated for more complete sequencing so that their genomes can serve as references for additional analysis, will be subjected to metabolic profiling, and possibly used as candidates for future whole-genome sequencing. From our collection of fungal isolates, we have so far submitted 13 pyrophilous fungi genomes and 11 transcriptomes as part of a JGI community sequencing project. Two genomes, *Crassisporium funariophilum* and *Pholiota highlandensis*, have been

assembled and annotated, and three more genomes are currently in sequencing. We are also in the process of growing a dominant pyrophilous fungal species, *Pyronema omphalodes*, on forest soil treated by a controlled burn to collect transcriptomic and metabolic data at individual time points, temperature profiles and soil depths.

*Objectives 2 and 3.* Temporal patterns in the degradation of PyOM and its sub-fractions will be assessed by extracting  $^{13}\text{C}$ -labelled polar and non-polar compounds and adding them to unlabeled char particles prior to incubation with soils. In order to trace degradation of each fraction, we have built an automated custom gas-sampling “multiplexer” connected to a cavity ringdown spectrometer, through which we can measure the efflux and isotopic composition of respired  $\text{CO}_2$ . The  $^{13}\text{C}$  labelled PyOM for these experiments is being produced from pine biomass grown in a labeling chamber constructed for this project. The biomass will be charred in a custom-built “charcoalator”, which allows for highly-controlled production of PyOM. The PyOM can be pyrolyzed over a range of temperatures, consistent with the physicochemical gradient model that evokes the steep heat gradient produced by forest fires to predict the effects of fire on the chemical properties of soils and biomass post-fire.

## References

1. Westerling, A.L., H.G. Hidalgo, D.R. Cayan, and T.W. Swetnam, Warming and earlier spring increase western US forest wildfire activity. *Science*, 2006. 313(5789): p. 940-943.
2. Gonzalez-Perez, J.A., F.J. Gonzalez-Vila, G. Almendros, and H. Knicker, The effect of fire on soil organic matter - a review. *Environment International*, 2004. 30(6): p. 855-870.
3. Santin, C., S.H. Doerr, C.M. Preston, and G. Gonzalez-Rodriguez, Pyrogenic organic matter production from wildfires: a missing sink in the global carbon cycle. *Global Change Biology*, 2015. 21(4): p. 1621-1633.
4. Lehmann, J., J. Skjemstad, S. Sohi, J. Carter, M. Barson, P. Falloon, K. Coleman, P. Woodbury, and E. Krull, Australian climate-carbon cycle feedback reduced by soil black carbon. *Nature Geoscience*, 2008. 1(12): p. 832-835.

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