

## Understanding Microbial Stress Responses in Soil Using Metagenome and Metatranscriptome Analysis

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**Project Goals:** Microorganisms play key roles in soil carbon turnover and stabilization of persistent organic matter via their metabolic activities, cellular biochemistry, and extracellular products. Microbial residues are the primary ingredients in soil organic matter (SOM), a pool critical to Earth's soil health and climate. We hypothesize that microbial cellular-chemistry, functional potential, and ecophysiology fundamentally shape soil carbon persistence, and we will characterize this via stable isotope probing (SIP) of genome-resolved metagenomes. We focus on soil moisture as a 'master controller' of microbial activity and mortality, since altered precipitation regimes are predicted across the temperate U.S. *Our SFA's ultimate goal is to determine how microbial soil ecophysiology, population dynamics, and microbe-mineral-organic matter interactions regulate the persistence of microbial residues under changing moisture regimes.*

Our SFA project objectives include:

- 1) Apply SIP-metagenomics to delineate how changing water regimes shape activity of individual microbial populations and expression of ecophysiological traits that affect the fate of microbial and plant C.
- 2) Identify and quantify mechanisms of mortality in the soil microbiome (focusing on phage lysis and water stress) and their contribution to C turnover and the biochemistry of microbial residues.
- 3) Measure how the soil microbiome and its products (cell envelope, extracellular polymeric substances, exo-enzymes) interact with contrasting mineral assemblages to control both short- and long-term soil C persistence.
- 4) Synthesize genome-scale ecophysiological trait data, population-specific growth and mortality, and SOM chemistry to build models of microbial functional guilds and SOM turnover, to predict the long-aspired connection between soil microbiomes and fate of soil C.

Managing soil health requires a detailed knowledge of how soil microbial metabolism and ecology affect soil organic matter formation and decomposition and respond to changes in the environment. It is often speculated that microbes in soil are C limited, either because of low C availability or low quality of the soil organic matter. This condition is thought to result in low C Use Efficiency, which reduces the microbial biomass and necromass production, with potentially negative effects on soil organic matter formation. Although we conceptually

understand how stress can affect soil organic matter formation, there are few direct ways to study stress in soil ecosystems. Here we propose to study microbial stress by analyzing the (meta)genome and (meta)transcriptome of bacterial species in soil ecosystems under high and low C availability.

Similar to “higher” organisms, bacteria have sophisticated response mechanisms to their environment. In this study, we will focus on bacterial sigma factors, transcription factors and regulators that are involved in changing gene-expression in response to stress conditions. We hypothesize that, by identifying transitions that results in the relative abundance of sigma factors, transcription factors and regulators, we can identify periods of microbial stress and measure how stress affects microbial C use efficiency, growth rates and overall community C and N cycling, and thus achieve greater insight into the processes that govern soil health.

We analyzed 10,601 bacterial genome, 4,929 metagenome and 1,753 metatranscriptome datasets from a wide range of ecosystems. Results show that of the various sigma factors, especially sigma E (regulating gene-expression in response to microbial envelope stress) is present at high relative abundance. A comparison between metagenomes and metatranscriptomes reveals that sigma factors associated with stress are less abundant in the metatranscriptomes than expected according to their abundances in metagenomes. This is interpreted to mean that microbial stress is not a dominant characteristic of microbial functioning in these environmental samples.