From natural carbon to a defined medium: Characterization of carbon utilization of FRC isolates

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Project Goals: ENIGMA -Ecosystems and Networks Integrated with Genes and Molecular Assemblies use a systems biology approach to understand the interaction between microbial communities and the ecosystems that they inhabit. To link genetic, ecological, and environmental factors to the structure and function of microbial communities, ENIGMA integrates and develops laboratory, field, and computational methods.

Natural organic matter (NOM) plays important roles in biological, chemical, and physical processes within the terrestrial ecosystem. The water-soluble fraction of NOM, known as dissolved organic matter (DOM), is associated with high microbial activity and respiration. Improving our knowledge of DOM composition is therefore critical to understand the native substrates used to support soil and sediment microbial communities.

Previously, we analyzed DOM of a saprolite soil collected from the Oak Ridge Field Research Center (ORFRC) using untargeted characterization of water-soluble soil microbial metabolites [1]. With this approach, 96 metabolites were identified, including amino acids, sugars, mono- and di-carboxylic acids, nucleobases, and nucleosides. This pool of metabolites was used to build a soil defined medium (SDM). Although SDM is environmentally based, it only supported the growth of only half of the tested bacterial from the ORFRC field site [1] which we now attribute to elemental stoichiometry especially the low carbon to nitrogen ratio which was 1. Here, we describe the construction of a new defined medium, NLDM, which additionally accounts for soil elemental stoichiometry, soil microbe metabolite use, and the composition of the widely-used R2A rich medium. We found that NLDM supported the growth of 53 of 53 phylogenetically diverse isolates we tested and enabled rapid profiling of substrate use via LC-MS/MS. We anticipate that this media may have additional use in isolating soil and sediment microorganisms.

Using ultrahigh resolution mass spectrometry, we were able to characterize ORFRC sediment DOM and its transformation by indigenous microbes. Lignin-like carbon, is the major component of DOM along the entire depth of sediment core. Labile carbon such as carbohydrate and amino sugar-like compounds are present in DOM from shallow surficial sediment. Decrease in abundance in deeper sediment, which happens to be more abundant in mainly recalcitrant carbon such as lipid and condensed aromatics. The dissolved organic nitrogen (or protein-like compounds) fraction of sediment DOM is an alternative carbon source for microbes inhabiting ORFRC groundwater when labile carbon is limited [2]. With sediment DOM as sole carbon source we successfully enriched rarely cultured phyla including *Verrucomicrobia*, *Planctomycetes*, and *Armatimonadetes*, and obtained isolates of several novel and undescribed organisms [3].

With our novel understanding of DOM and their transformations by FRC isolates using NLDM are now being used to link our lab studies to field observations. This will result in a better understanding of the coupling between growth substrates, other environmental controls, and microbial community activity and structure.

References

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