How Monomer Availability Impacts the Scramble for Carbon During Plant Polymer Decomposition: Using Pure Cultures as a Predictive Model System.

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Project Goals: Microbes are responsible for decomposing plant materials in terrestrial systems, which has an important impact on carbon sequestration. During the decomposition of plant materials, microorganisms take up monomers for use in respiration and biosynthesis. Plant cell wall polymers, such as cellulose, pectin and lignin, are degraded into monomers by microorganisms capable of producing extracellular enzymes. However, production of extracellular enzymes is an energy-intensive process. Therefore, our project goal is to describe in detail how monomer availability affects microbial physiology and gene transcription. Furthermore, from this information we will predict the identity and importance of "cheating" organisms during decomposition. In this project, we categorize microorganisms involved in decomposition as 1) Investors, the microorganisms that produce extracellular enzymes to break down complex plant material into simple molecules; 2) Obligate cheaters, who do not carry genes of extracellular enzymes but who take up monomers released from plant polymers; 3) Opportunistic cheaters, who do have genes of extracellular enzymes, but suppress expression of these enzymes and still take up degradation products.

In order to understand the interaction between decomposer activity and carbon supply during the decomposition process, we grew *Talaromyces stipitatus* NRRL 1006, a potential investor/opportunistic cheater, in sand microcosms with cellulose as the sole carbon source. We amended microcosms with monomers in two experiments to monitor shifts in gene expression and microbial physiology in response to different amounts and types of labile carbon monomers. Here we describe patterns in respiration; responses in extracellular enzyme activity, biomass, and gene expression will also be presented. In the first experiment, xylose was amended at four concentrations ranging from 10 mM to 120 mM in order to examine concentration thresholds resulting in a physiological and transcriptomic response to monomers. Respiration responded to all xylose amendments, but was highest for the 60 and 120 mM amendments. In the second experiment, we amended microcosms with 60 mM solutions of glucose, xylose, galacturonic acid, and vanillin to determine the effect of switching from growth on cellulose and corresponding glucose monomers, to other monomer types. Surprisingly, respiration increased the most due to xylose amendment, followed by glucose and galacturonic acid. Respiration was suppressed by vanillin.

These experiments will be repeated with 13 additional model fungal and bacterial species with completed genomic sequences. These organisms include potential investors and cheaters. In initial experiments, the respiration rate of investors growing on cellulose in sand microcosms showed periodic increases and decreases during 20 days incubation, whereas the respiration rate of cheaters peaked quickly and then went flat. Small subunit RNA copy numbers of cheaters and potential investors continued to shift following leveling off of respiration rate.

Combining data from these experiments, it is hypothesized that “cheaters” will be identifiable as the organisms with the highest monomer concentration thresholds resulting in suppression of extracellular enzyme genes.

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