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Using RTMS and IRMS to Unravel the Effect of Nitrogen on the Microbiome Molecular Mechanisms of Carbon and Nutrient Cycling during Rewetting after Drought

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Soil is a highly diverse ecosystem where water availability can drive microbial cell function and influence in situ substrate fate and carbon cycling. Hydration levels can thus direct carbon and nutrient cycles in the microbiome in these soils. The rapid carbon release and nutrient utilization upon hydration of desiccated soils has been observed for decades, but little is known about the controls and timing of the underlying molecular events. This research is aimed at mapping the interdependence of carbon and associated nutrient cycling in soils using ^{13}C labeled substrates as a tracer of substrate fluxes in desiccated soil microbiomes after rewetting in combination with our novel Real Time Mass Spectrometry (RTMS) system and isotope ratio mass spectrometry (IRMS).

In our experiments, soil amended with two different nitrogen sources was exposed to a 2 week drought then supplemented with ^{13}C glucose solution to mimic a rewetting event. The RTMS traced the production of $^{12}CO_2$ and $^{13}CO_2$ every 5 sec for 24 hours after the addition of water. We observed that the rate of $^{12}CO_2$ production from all the samples within the first 15 minutes after rewetting were similar, indicating that the initial burst of CO_2 is not nitrogen dependent. The overall amount of CO_2 produced differed based on the type of nitrogen supplied, with CO_2 production being lowest with alanine supplementation. Using ^{13}C labeled alanine as a sole carbon source (amended into the rewetting solution) we found that the amino acid is respired very slowly and is not extensively used as an energy source. We hypothesize that the carbon from the glucose was diverted to biosynthetic pathways rather than energy production when in the presence of an abundance of the amino acid. We will validate the utilization of the carbon from the amended glucose into synthesized proteins using IRMS analyses of the $^{12}C/^{13}C$ ratio in metabolites, proteins, and lipid fractions. these studies will help unravel the molecular mechanisms underlying the Birch Effect first observed more than 50 years ago and known to influence terrestrial carbon cycling.