Field Measurements of Taxon-Specific Microbial Growth in Soil at Two Elevation Gradient Sites Using Quantitative Stable Isotope Probing (qSIP)

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Project Goals: The goal of this study was to determine environmentally relevant measurements of microbial growth using a quantitative stable isotope probing technique with ^{18}O-H_2O. Growth in intact soils in the field was compared with laboratory-based soil incubations, and the response of microbial growth to warming along an elevation gradient was determined.

Measuring microbial growth in environmental samples remains challenging. Determining what microbes are present, their activity, and rates of activity and growth is critical to inform their role in element and nutrient cycling, ecosystem function, and responses to environmental change. Attempts to measure *in situ* microbial cell replication include stable isotopically labeled substrate (^{13}C,^{18}O) enrichment into DNA, algorithms based on metagenome and genome sequence coverage, cell enumeration with microscopy or flow cytometry, and ^{3}H and ^{14}C substrate incorporation into cells. Many techniques, especially for substrate incorporation experiments, rely on laboratory incubations where the sample becomes highly disturbed and perhaps might not capture activity accurately. Here, we aimed to measure *in situ* microbial taxon-specific growth in disturbed and undisturbed soil as well as growth in warmed soil cores. Field and laboratory-based incubations for quantitative stable isotope probing (qSIP) with ^{18}O-H_2O were conducted to determine microbial growth. qSIP involves a combination of techniques involving CsCl density gradient separation, quantitative PCR to determine 16S rRNA gene copy number, and 16S rRNA gene sequencing, all combined to obtain microbial taxa specific growth and death. ^{18}O-H_2O was injected into undisturbed soil in a temperate mixed conifer soil and a ponderosa pine forest soil, two sites part of the C. Hart Merriam Elevation Gradient in Northern Arizona. The soil was left to incubate at field conditions for three and ten days. Parallel ten-day incubations from the mixed conifer soil were incubated in the laboratory to compare microbial taxa growth in disturbed soil incubations versus field conditions. This study, using the novel qSIP technique in a field incubation will begin to elucidate potential laboratory artifacts in studying microbial activity and give more environmentally relevant measures of microbial replication. Preliminary results of this experiment will be discussed.

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