

Spatially-resolved carbon flow through a hypersaline microbial mat

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Hot Lake is a hypersaline, meromictic lake located in a closed basin in north-central Washington. Salinity in Hot Lake is driven by magnesium sulphate (up to 2 M) and despite this extreme salinity, thick (up to 4 cm) laminated, phototrophic, benthic mat develops seasonally.

We used a suite of approaches to track ¹³C labelled substrates into the mat community to probe how metabolic interactions impart system-level properties. We performed a series of *ex situ* incubations over a diel cycle using fresh mat samples in lake water amended with ¹³C-labeled bicarbonate, acetate, or glucose. Bulk mat stable isotope analysis quantified uptake of these substrates into the mat and showed net biomass increase during daylight and carbon loss during the night. Laser ablation IRMS enabled a spatially resolved localization of label accumulation within mat cross sections and permitted tracking of subsequent label migration. Intracellular metabolomics identified osmolytes (including glucose and trehalose) as having very high turnover in the system. Interestingly, different intramolecular labelling patterns were observed in compounds such as 3-hydroxybutanoic acid and 3-hydroxypentanoic acid, suggesting different biochemical processing of added acetate and glucose. Protein stable isotope probing showed that phototrophic species most rapidly accumulated labelled bicarbonate in their biomass.