# From Position-Specific Labeling to Environmental Fluxomics: Elucidating Biogeochemical Cycles from the Metabolic Perspective (BG Division Outstanding ECS Award Lecture) 

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#### Abstract

Understanding soil and sedimentary organic matter (SOM) dynamics is one of the most important challenges in biogeoscience. To disentangle the fluxes and transformations of C in soils a detailed knowledge on the biochemical pathways and its controlling factors is required. Biogeochemists' view on the C transformation of microorganisms in soil has rarely exceed a strongly simplified concept assuming that C gets either oxidized to $\mathrm{CO}_{2}$ via the microbial catabolism or incorporated into biomass via the microbial anabolism. Biochemists, however, thoroughly identified in the past decades the individual reactions of glycolysis, pentose-phosphate pathway and citric acid cycle underlying the microbial catabolism. At various points within that metabolic network the anabolic fluxes feeding biomass formation branch off. Recent studies on metabolic flux tracing by position-specific isotope labeling allowed tracing these $C$ transformations in soils in situ, an approach which is qunatitatively complemented by metabolic flux modeling. This approach has reached new impact by the cutting-edge combination of position-specific 13C labeling with compound-specific isotope analysis of microbial biomarkers and metabolites which allows 1) tracing specific anabolic pathways in diverse microbial communities in soils and 2) identification of specific pathways of individual functional microbial groups. Thus, the combination of position-specific labeling, compound-specific isotope incorporation in biomarkers and quantitative metabolic flux modelling provide the toolbox for quantitative soil fluxomics.


Our studies combining position-specific labeled glucose with amino sugar 13C analysis showed that up to $55 \%$ of glucose, incorporated into the glucose derivative glucosamine, first passed glycolysis before allocated back via gluconeogenesis. Similarly, glutamate-derived C is allocated via anaplerotic pathways towards fatty acid synthesis and in parallel to its oxidation in citric acid cycle. Thus, oxidizing catabolic pathways and anabolic pathways, i.e. building-up new cellular compounds, occurred in soils simultaneously, a combination unlikely to occur in pure cultures, where constant growth conditions under high C supply allow a straight unidirectional regulation of C metabolism. However, unstable environmental conditions, C scarcity and interactions between a still unknown diversity of microorganisms in soils are likely to induce the observed metabolic diversity. Coupling these results with the position-specific fingerprint of microbial biomarkers revealed that microbial groups show deviating adaptation strategies and that they react on environmental changes by activation or deactivation of specific metabolic pathways such as anaplerotic fluxes.

To understand how microorganisms catalyze the biogeochemical fluxes in soil a profound understanding of their metabolic adaptation strategies such as recycling or switching between pathways is crucial. Metabolic flux models adapted to soil microbial communities and their regulatory strategies will not only deepen our understanding on the microorganims' reactions to environmental changes but also create the prerequisits for a quantitative prediction of biogeochemical fluxes based on the underlying microbial processes.

