**Quantitative assessment of biodegradation in the subsurface using stable isotope analysis – theoretical assessment and reactive transport simulations**

Martin Thullner, Falk Heße and Florian Centler

Department of Environmental Microbiology, Helmholtz Centre for Environmental Research – UFZ, Leipzig, Germany

Many organic compounds including contaminants are subject to microbial degradation in the subsurface. A reliable quantification of such in situ degradation processes is still a major challenge. Compound specific stable isotope analysis has become an established method for the qualitative analysis of biodegradation in the field and this method is also proposed for a quantitative analysis. However, in-situ conditions are characterized by a complex interplay between the degradation process and transport processes at different scales. These transport processes range from dispersive mixing at large scales to pore-scale mass transfer controlling the availability of compounds to the degrading microorganisms. A straightforward application of simplified analyses of stable isotope data may thus lead to misleading quantifications of the investigated degradation process.

Furthermore, microbial degradation of organic compounds may occur via different redox-specific degradation pathways. At sites with varying redox conditions, different degradation pathways can contribute to total compound degradation. The identification of the individual pathway-specific contributions to total compound removal is of high practical relevance especially in case of contaminants, but such quantitative assessment is difficult to achieve with available methods. Current stable-isotope-fractionation-based analyses focus on the identification of the dominant biodegradation pathway under constant environmental conditions while an extension of such approaches is needed in case of several degradation pathways providing a significant contribution to total degradation.

We discuss the limitations of such simplified analyses methods and present a set of theory-derived extensions which allow considering transport-derived effects for obtaining more reliable quantitative estimates of degradation based on stable-isotope data. Furthermore, we present an approach based on multiple stable isotope data to estimate the individual contributions of different redox specific pathways.

To test the applicability of these approaches at in-situ conditions, we performed numerical reactive transport simulations at the pore-scale and at the macro-scale. Model simulations consider the advective-dispersive transport of dissolved species, their biodegradation via one or several different redox-specific degradation pathways, and the fractionation of stable isotopes caused by the individual degradation pathways. Results confirm the general applicability of the presented estimation methods and allow determining their range of application. The presented methods complement the stable-isotope-fractionation-based analysis toolbox.