



Short-term transformation of alanine in soil assessed by position-specific labeling

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Transformation of low molecular weight organic substances (LMWOS) in soil is one of the most important processes in the turnover of organic matter as all high molecular substances pass the stage of LMWOS during decomposition. We suggested using a unique feature of isotope applications – the position-specific labeling – to get a closer look on the mechanisms of LMWOS transformation in soil. This tool allows to distinguish the flux of carbon from individual positions in the molecule and tracing of individual C atoms in all metabolites within any pools.

We assessed short-term turnover of C from different positions of the amino acid alanine: Uniformly and position-specifically labeled ^{14}C -alanine were mixed with soil, centrifuged and ^{14}C activity was measured at increasing time steps. Three sterilization treatments of the soil allowed to differentiate between the potential mechanisms of a declining ^{14}C activity in soil solution: i) sorption, exoenzyme activity and microbial uptake of alanine from the unsterilized soil; ii) only sorption and exoenzymatic degradation was possible by inhibition of the intracellular metabolism by NaN_3 , iii) whereas in treatment with total protein denaturation by HgCl_2 , sorption was the only process removing ^{14}C activity from the supernatant. In addition to alanine in solution, alanine sorbed to soil minerals (hematite, goethite, smectite, kaolinite and active coal) was added to non-sterilized soil and degradation of the sorbed amino acid was observed by $^{14}\text{CO}_2$.

Sorption mainly occurred as a whole molecule, whereas C from different positions of alanine showed different patterns of exoenzymatic degradation and microbial utilization. The carboxyl group was mainly affected by microbial decomposition and most of these processes occurred in microbial cells and not by exoenzymes. In contrast the CH_3 -position of the amino acid was mainly incorporated into microbial biomass and were less decomposed to CO_2 . We showed that C from different positions of alanine has differing fate in soil, even in short-term experiments.

Further factor with considerable influence on the fate of LMWOS in soil is their concentration. We simulated the concentration i) typical for hot spot nearby dying root cells with a $500 \mu\text{M}$ alanine solution and ii) conditions typical nearby degraded proteinogenous plant material with a $50 \mu\text{M}$ alanine solution. We used a iii) $5 \mu\text{M}$ concentration as this is the mean concentration in soil solution and iv) $0.5 \mu\text{M}$ concentration for simulating root free soil. Under LMWOS concentrations sufficient for microbial growth (50 and $500 \mu\text{M}$) significant differences in the position-specific utilization of alanine C could be observed, whereas low concentrations of alanine (0.5 and $5 \mu\text{M}$) lead to an intact uptake and microbial utilization of all three C positions.

Desorption of the LMWOS revealed to which extent alanine is bound by ion exchange, ligand exchange or is irreversibly bound to the mineral surfaces. In non-sterilized treatments mainly the transformation products of alanine were desorbed from the soil surface. The addition of sorbed alanine to soil shows, that the type of adsorbing mineral surface has a great influence on the position-specific degradation of LMWOS.

We showed that the application of position-specifically labeled substances opens a new way to investigate the LMWOS-transformations in soil. The transformations of single C atoms allow conclusions about the individual transformation steps, their rates on a submolecular level and improve our understanding of soil carbon fluxes.