



## **Utilization of low molecular weight organics by soil microorganisms: combination of $^{13}\text{C}$ -labelling with PLFA analysis**

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Microbial metabolism is the main transformation pathway of low molecular weight organic substances (LMWOS), but detailed knowledge concerning the fate of LMWOS in soils is strongly limited. Considering that various LMWOS classes enter biochemical cycles at different steps, we hypothesise that the percentage of their LMWOS-Carbon (C) used for microbial biomass (MB) production and consequently medium-term stabilisation in soil is different.

We traced the three main groups of LMWOS: amino acids, sugars and carboxylic acids, by uniformly labelled  $^{13}\text{C}$ -alanine, -glutamate, -glucose, -ribose, -acetate and -palmitate. Incorporation of  $^{13}\text{C}$  from these LMWOS into MB (fumigation-extraction method) and into phospholipid fatty acids (PLFAs) (Bligh-Dyer extraction, purification and GC-C-IRMS measurement) was investigated under field conditions 3 d and 10 d after LMWOS application. The activity of microbial utilization of LMWOS for cell membrane construction was estimated by replacement of PLFA-C with  $^{13}\text{C}$ .

Decomposition of LMWOS-C comprised 20–65% of the total label, whereas incorporation of  $^{13}\text{C}$  into MB amounted to 20–50% of initially applied  $^{13}\text{C}$  on day three and was reduced to 5–30% on day 10. Incorporation of  $^{13}\text{C}$ -labelled LMWOS into MB followed the trend sugars > carboxylic acids > amino acids. Differences in microbial utilisation between LMWOS were observed mainly at day 10. Thus, instead of initial rapid uptake, further metabolism within microbial cells accounts for the individual fate of C from different LMWOS in soils.

Incorporation of  $^{13}\text{C}$  from each LMWOS into each PLFA occurred, which reflects the ubiquitous ability of all functional microbial groups for LMWOS utilization. The preferential incorporation of palmitate can be attributed to its role as a direct precursor for many fatty acids (FAs) and PLFA formation. Higher incorporation of alanine and glucose compared to glutamate, ribose and acetate reflect the preferential use of glycolysis-derived substances in the FAs synthesis pathway.

Gram-negative bacteria (16:1w7c and 18:1w7c) were the most numerous and active in using LMWOS. Their high activity corresponds to a high demand for anabolic products, leading to an increased incorporation of pentose-phosphate pathway C i.e. ribose-C into PLFAs. The turnover of  $^{13}\text{C}$ -sugar and  $^{13}\text{C}$ -amino acid in filamentous microorganisms was lower than in all procaryotic groups. However, their turnover was in the same range as that of gram-positive bacteria if  $^{13}\text{C}$ -carboxylic acids were considered as the substrate. This is associated with the preference of fungi or filamentous microorganisms in general, for acidic and more complex organics.

Thus, we showed the divergence of C pathways from LMWOS over the medium-term, despite their similar initial uptake by microorganisms. Further investigation of the formation of microbial compounds from LMWOS and their stabilisation in soils is necessary to improve our understanding of the impact of this highly available C on the soil C cycle.