



## **Utilisation of young and old soil carbon sources by microbial groups differ during the growing season and between experimental treatments in a long-term field experiment**

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Soil organic matter (SOM) is the largest active carbon pool in the terrestrial environment. SOM is a key factor for soil fertility, but is also important for the sequestration of atmospheric CO<sub>2</sub>. In agricultural soils, management of plant residues and the use of organic fertilisers play important roles for maintaining SOM. Switching from C3 plants to C4 plants such as maize, enables a natural labelling in situ; when coupled with compound specific <sup>13</sup>C isotope analysis of phospholipid fatty acids (PLFAs) it allows the proportion of new C (fixed after the switch added to soil from above- and belowground litter and root exudates) and the proportion of old C (fixed prior to the switch derived from turnover of organic matter) utilised by the soil microbial community to be determined. (new paragraph) A field experiment in Sweden, amended with different mineral and organic fertilisers since 1956, was grown with C3 plants, mainly cereals until 1999. From the year 2000 silage maize was grown every year. In 2012, soil from four replicate plots of five experimental treatments, N fertilised, N fertilised amended with straw and sewage sludge, and two controls (bare fallow and cropped unfertilised) were sampled three times, at the start, middle and end of the growing season. Phospholipid fatty acids (PLFAs) were extracted from all soil samples and analysed for concentrations and <sup>13</sup>C content. (new paragraph) Total PLFA concentrations and also the PLFA/SOM ratios increased with SOM in the different treatments. Seasonal variation in total PLFA was small except for the most SOM-rich treatment (sewage sludge) where concentrations significantly decreased during the growing season indicating the depletion of a labile SOM pool. Weighted mean values of  $\delta^{13}\text{C}$  in PLFAs show that the plots fertilised with only calcium nitrate had the highest  $\delta^{13}\text{C}$ -values in PLFAs before (-20.24 ‰) and after the vegetation period (-20.37 ‰), due to a large input of <sup>13</sup>C-enriched plant material. However, during the vegetation period the values were much lower (-21.85 ‰). This coincided with a strong increase of the PLFA 18:2 (from 0.99 up to 2.37 nmol g dry wt soil<sup>-1</sup>), indicating utilisation of old organic matter by fungi, while mono-unsaturated PLFAs, indicating Gram-negative bacteria, were more frequent before and after the growing season. Microbial dynamics in the unfertilised control followed the same seasonal pattern but PLFAs were less enriched in <sup>13</sup>C due to lower yields compared with the N-fertilised treatment. The addition of organic amendments (straw or sewage sludge) lowered  $\delta^{13}\text{C}$ -values in PLFAs below values of the control due to input of labile material with C3-origin. PLFAs in the bare fallow treatment, that had not received plant carbon inputs during twelve years, were most <sup>13</sup>C depleted among the treatments but still enriched by about 2‰ compared with SOM, indicating a degree of microbial fractionation.