

## **Microbial metabolism in soil at low temperatures: Mechanisms unraveled by position-specific $^{13}\text{C}$ labeling**

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Microbial transformation of organic substances in soil is the most important process of the C cycle. Most of the current studies base their information about transformation of organic substances on incubation studies under laboratory conditions and thus, we have a profound knowledge on SOM transformations at ambient temperatures. However, metabolic pathway activities at low temperature are not well understood, despite the fact that the processes are relevant for many soils globally and seasonally.

To analyze microbial metabolism at low soil temperatures, isotopomers of position-specifically  $^{13}\text{C}$  labeled glucose were incubated at three temperature; 5, -5 -20 oC. Soils were sampled after 1, 3 and 10 days and additionally after 30 days for samples at -20 °C. The  $^{13}\text{C}$  from individual molecule position was quantified in respired  $\text{CO}_2$ , bulk soil, extractable organic C and extractable microbial biomass by chloroform fumigation extraction (CFE) and cell membranes of microbial communities classified by  $^{13}\text{C}$  phospholipid fatty acid (PLFA) analysis.

$^{13}\text{CO}_2$  released showed a dominance of the flux from C-1 position at 5 °C. Consequently, at 5 °C, pentose phosphate pathway activity is a dominant metabolic pathway of glucose metabolization. In contrast to -5 °C and -20 oC, metabolic behaviors completely switched towards a preferential respiration of the glucose C-4 position. With decreasing temperature, microorganism strongly shifted towards metabolization of glucose via glycolysis which indicates a switch to cellular maintenance. High recoveries of  $^{13}\text{C}$  in extractable microbial biomass at -5 °C indicates optimal growth condition for the microorganisms. PLFA analysis showed high incorporation of  $^{13}\text{C}$  into Gram negative bacteria at 5 °C but decreased with temperature. Gram positive bacteria out-competed Gram negatives with decreasing temperature. This study revealed a remarkable microbial activity at temperatures below 0 °C, differing significantly from that at ambient temperatures. These metabolic pathways, can be unraveled based on position-specific labeling.