



## Distinct fungal and bacterial $\delta^{13}C$ signatures can drive the increase in soil $\delta^{13}C$ with depth

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Soil microbial biomass is a key precursor of soil organic carbon (SOC), and the enrichment in  $^{13}C$  during SOC diagenesis has been purported to be driven by increasing proportions of microbially derived SOC. Yet, little is known about how the  $\delta^{13}C$  of soil microbial biomass - and by extension the  $\delta^{13}C$  of microbial inputs to SOC - vary in space, time, or with the composition of the microbial community. Phospholipid fatty acids (PLFA) can be analyzed to measure the variation of the natural abundance  $\delta^{13}C$  values of both individual groups of microorganisms and the microbial community as a whole. Here, we show how variations of  $\delta^{13}C_{PLFA}$  within the soil profile provides insight into C fluxes in undisturbed soils and demonstrate that distinct  $\delta^{13}C$  of fungal and bacterial biomass and their relative abundance can drive the increase of bulk  $\delta^{13}C_{SOC}$  with depth.

We studied the variation in natural abundance  $\delta^{13}C$  signatures of PLFA in podzolic soil profiles from mesic boreal forests in Atlantic Canada. Samples from the organic horizons (L,F,H) and the mineral (B; top 10 cm) horizons were analyzed for  $\delta^{13}C$  values of PLFA specific to fungi, G+ bacteria, or G- bacteria as proxies for the  $\delta^{13}C$  of the biomass of these groups, and for  $\delta^{13}C$  values of PLFA produced by a wide range of microorganisms (e.g. 16:0) as a proxy for the  $\delta^{13}C$  value of microbial biomass as a whole. Results were compared to fungi:bacteria ratios (F:B) and bulk  $\delta^{13}C_{SOC}$  values.

The  $\delta^{13}C$  values of group-specific PLFA were driven by differences among source organisms, with fungal PLFA consistently depleted (2.1 to 6.4‰) relative to and G+ and G- bacterial PLFA in the same sample. All group-specific PLFA, however, exhibited nearly constant  $\delta^{13}C$  values throughout the soil profile, apparently unaffected by the over 2.8‰ increase in  $\delta^{13}C_{SOC}$  with depth from the L to B horizons. This indicates that bulk SOC poorly represents the substrates actually consumed by soil microorganisms in situ. Instead, our results suggest that soil microorganisms primarily consume substrates that exhibit constant  $\delta^{13}C$  values throughout the soil profile, like root litter or dissolved organic carbon from litter leachates or root exudates that percolates through the soil column.

$\delta^{13}C$  values of PLFA produced by both fungi and bacteria, in contrast to the group specific PLFA, strongly increased with depth and were tightly correlated to F:B ratios ( $R^2 > 0.84$ ), which decreased with depth. Because group-specific PLFA did not exhibit increased  $\delta^{13}C$  with depth, the increase observed in the general biomarker  $\delta^{13}C$  values, associated with the aggregated microbial community, was not the consequence of microbial incorporation of more  $^{13}C$  enriched SOC at greater depth. Rather, the increase in community  $\delta^{13}C$  reflects a shift in community structure towards more  $^{13}C$  enriched bacteria with depth. Our results indicate that, higher  $\delta^{13}C$  values associated with microbial biomass at a greater depth likely contributes to the increase in  $\delta^{13}C_{SOC}$  with depth via more  $^{13}C$  enriched contributions from necromass to SOC.