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Molecular multiproxy analysis of ancient root systems suggests strong alteration of deep subsoil organic matter by rhizomicrobial activity

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Roots have a high potential capacity to store large amounts of CO_2 in the subsoil. However, associated with rooting, microorganisms enter the subsoil and might contribute to priming effects of carbon mineralisation in the microbial hotspot rhizosphere. Although these processes are well known for recent surface soils, it remains questionable, if and how microorganisms contribute to priming effects in the subsoil and if these effects can be traced after the roots‘ lifetime. The current study implies several state-of-the-art techniques like DNA and lipid molecular proxies to trace remains of microbial biomass in ancient root systems. These can provide valuable information if parts of the root and rhizomicrobial biomass are preserved, e.g. by encrustation with secondary carbonate during the root's lifespan or shortly thereafter.

At the Late Pleistocene loess-paleosol sequence near Nussloch (SW Germany), rhizoliths (calcified roots) occur highly abundant in the deep subsoil from 1 to 9 m depth and below. They were formed by Holocene woody vegetation. Their size can account for up to several cm in diameter and up to $>$ 1 m length. Rhizoliths and surrounding sediment with increasing distances of up to 10 cm, as well as reference loess without visible root remains were collected at several depth intervals. Samples were analysed for n-fatty acids (FAs) and glycerol dialkyl glycerol tetraethers (GDGTs; membrane lipids from Archaea and some Bacteria), as well as structural diversity based on the RNA gene of the prokaryotic ribosome subunit 16S (16S rRNA). GDGT represent organic remains from microbial biomass, whereas FA comprise both microbial remains and degradation products. 16S rRNA indicates the presence of both living cells and/or cell fragments.

Despite the general low RNA contents in the sample set, results pointed to a much higher abundance of bacterial compared to archaeal RNA. The latter occured in notable amounts only in some rhizoliths. This was in part enforced by decreasing contents of archeal GDGTs from rhizolith via rhizosphere towards root-free loess. Furthermore, the bacterial fingerprint revealed – similar to modern root systems – higher taxonomic diversity in rhizosphere compared to rhizoliths and reference loess. This argues for microorganisms benefiting from root deposits and exudates. Highest concentrations of branched GDGTs in rhizoliths suggest that their source organisms feed on root remains. Incorporation of rhizomicrobial remains as represented by RNA and GDGTs usually affected the sediment at maximum to a distance of 2-3 cm from the former root.

FA contents in rhizosphere showed strong scatter and were in part depleted compared to reference loess or, especially in deeper transects, enriched. This indicates the presence of degradation products originating from former rhizosphere processes. Especially at larger depth not affected by modern pedogenic processes, portions of mainly microbial derived C_{16} homologues were higher in rhizosphere loess up to distances of 10 cm, revealing that the possible extension of the rhizosphere was underestimated so far.

In C_{org} poor subsoil, the occurence of diverse rhizosphere microorganisms and degradation processes even in several centimeters distant from roots point to a strong alteration of OM, possibly contributing to carbon mineralisation.