Title: **Microbial Response to Root Substrate Addition is Depth Dependent**

Elaine Pegoraro,[1] Jennifer L Soong,[2] Cristina Castanha,[1] Cyrill Zosso,[3] Guido L.B. Wiesenberg,[3] Michael W.I. Schmidt,[3] and Margaret Torn[1]

[1]Lawrence Berkeley National Laboratory, Berkeley, CA.
[2]Colorado State University, Fort Colins, CO
[3]University of Zurich, Zurich, Switzerland

Contact: (elainepegoraro@lbl.gov)

Project Lead Principal Investigator (PI): Margaret Torn

BER Program: ESS

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Project Websites: https://eesa.lbl.gov/projects/terrestrial-ecosystem-science/

https://tes.lbl.gov/

Project Abstract:

The subsoil (<30 cm) contains almost half of the soil organic carbon (SOC) stock in the top meter of soil (Jobbágy & Jackson 2000, Balesdent et al. 2018), yet most studies only focus on surface layers (Yost & Hartemink 2020). Subsoil temperatures are projected to increase at a similar rate to air and surface soil temperatures (Soong et al. 2020), which can make this deep, relatively stable SOC pool vulnerable to increased microbial decomposition. Nonetheless, there is still no clear consensus on the factors that drive warming-induced changes in SOC stocks (van Gestel et al.2018). To understand the capacity of subsoil to store SOC with climate change, we must understand how decomposition changes with depth and warming.

We examined *in situ* litter decomposition by adding ^{13}C-labelled fine roots to surface (10-14 cm), mid- (40-44 cm), and deep soil layers (85-89 cm) of a coniferous temperate forest in soil classified as alfisols. At our site, we have warmed soil by 4°C relative to control temperatures to 100 cm since 2013. We recovered the root-derived C in bulk soil and microbial phospholipid fatty acids (PLFA) after 1 and 3 years of incubation. The overall recovered litter fraction in bulk soil significantly decreased by 60% in the surface and by 90% in the mid- and deep soil layers from 2017 to 2019, indicating that microbes in the subsoil relied on the added root-C more than surface microbes. Overall, total PLFA significantly decreased with depth (p-value <.0001), corroborating previous work at our site showing a significant decrease in microbial biomass with depth (Zosso et al. 2021). Though the root addition treatment did not significantly change total PLFA, it was on average higher at 85-89 cm relative to the disturbance control. Root addition significantly increased the relative abundance of fungi at 85-89 cm relative to the rest of the soil profile, contrasting previous findings that fungal biomarkers that relied on the added root substrate were less abundant at depth (Hicks Pries et al. 2018). Microbes in subsoil preferentially used the added substrate during the incubation period, leading to an increase in the fungal relative abundance at

depth. Our results suggest that particulate SOC in subsoil could be vulnerable to increased microbial decomposition, which can be exacerbated by warmer soil temperatures (Soong et al. 2021).

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