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Nanoscale STXM imaging of soil fungal exudates and organo-mineral interfaces

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Soils act as a major sink for atmospheric carbon (C) and, correctly managed, can help counterbalance the excessive CO₂ emissions. Organic C in soils can be physically stabilized and 'hidden' from its decomposers within soil aggregates and it is thought that soil fungi play a decisive role in "gluing together" and redistributing soil mineral particles and existing organic matter to form them (M. W. I. Schmidt et al., *Nature* 478(7367), 49–56, 2011). A significant contribution to the early aggregation process is adsorption of fungal exudates to the reactive surfaces of mineral particles. To uncover the mechanisms of C stabilization processes and to be able to increase the C sink potential of our soils, we need a deepened understanding of which fungi play key roles in the process, what mineral properties promote it, and what type of fungal exudates are involved.

For this purpose, we have grown saprotrophic and symbiotic (both arbuscular mycorrhizal (AM) and ectomycorrhizal (EM)) fungi under sterile conditions in contact with different principal soil components: quartz, goethite and muscovite, on top of X-ray transparent silicon nitride membrane windows and analyzed fungal hyphae by high lateral resolution synchrotron based scanning transmission X-ray microscopy (STXM) in combination with near edge X-ray fine structure (NEXAFS) spectroscopy at absorption edges of C(K), K(L), N(K) and Fe(L). We performed our experiments in the SM beamline at Canadian Light Source, Saskatoon, Canada and I08 beamline at Diamond Light Source, Oxfordshire, UK. In the resultant chemical images, we were able to differentiate the mostly proteinaceous hyphal material, the exudate layer constituting of mixtures of polysaccharides and proteins, and the organo-mineral interfaces consisting of a higher protein and carboxyl to sugar ratio than in the exudate layer. We also observed heterogeneous distributions of the exudate materials around the fungal hypha, indicating presence of exudation channels in the cell wall. Finally, we specifically analyzed NEXAFS spectra at Fe(L) absorption edge of goethite containing samples and were able to show changes in iron speciation in the mineral particles that were in contact with the fungal exudates. These results provide us with better insights to both nanoscale processes of fungal exudation and their role in the formation of organo-mineral interfaces subsequently responsible for soil aggregation.