

## Carbon transfer from plant roots to soil – NanoSIMS analyses of undisturbed rhizosphere samples

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Soils are composed of a wide diversity of organic and mineral compounds, interacting to form complex mosaics of microenvironments. Roots and microorganisms are both key sources of organic carbon (OC). The volume of soil around living roots, i.e. the rhizosphere, is a privileged area for soil microbial activity and diversity. The microscopic observation of embedded soil sections has been applied since the 1950's and has enabled observation of the rhizosphere at the smallest scale of organism interaction, i.e. at the level of root cells and bacteria (Alexander and Jackson, 1954). However, the observation of microorganisms in their intact environment, especially in soil, remains challenging. Existing microscopic images do not provide clear evidence of the chemical composition of compounds observed in the rhizosphere. Nano-scale secondary ion mass spectrometry (NanoSIMS) is a high spatial resolution method providing elemental and isotopic maps of organic and mineral materials. This technic has been increasingly used in soil science during the last decade (Hermann et al., 2007; Vogel et al., 2014) and more specifically for undisturbed soil sample observations (Vidal et al., 2016). In the present study, NanoSIMS was used to illustrate the biological, physical and chemical processes occurring in the rhizosphere at the microscale. To meet this objective, undisturbed rhizosphere samples were collected from a field experiment in Switzerland where wheat plants were pulse-labelled with 99%  $^{13}\text{C}$ -CO<sub>2</sub> in weekly intervals throughout the growing season and sampled at flowering. Samples were embedded, sectioned, polished and analyzed with NanoSIMS, obtaining secondary ion images of  $^{12}\text{C}$ ,  $^{13}\text{C}$ ,  $^{12}\text{C}^{14}\text{N}$ ,  $^{16}\text{O}$ ,  $^{31}\text{P}^{16}\text{O}_2$ , and  $^{32}\text{S}$ . The  $\delta^{13}\text{C}$  maps were obtained thanks to  $^{12}\text{C}$  and  $^{13}\text{C}$  images.  $^{13}\text{C}$  labelled root cells were clearly distinguished on images and presented highly variable  $\delta^{13}\text{C}$  values. Labelled spots ( $< 1 \mu\text{m}$ ), identified as bacteria, were located at the root cell surroundings. These microorganisms were intimately associated with soil particles, forming microaggregates tightly bound to root cells. Finally, some images revealed the presence of larger labelled spots ( $> 4 \mu\text{m}$ ) potentially assignable to arbuscular fungal hyphae. These results illustrate the transfer of carbon from the root tissues towards the microbial communities and the direct fate as organo-mineral associated OC at mineral soil particles.

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