Experimental report

Proposal:	UGA	-111	Council: 4/2020			
Title:	Comb	Combining neutron laminography with optical fluorescence imaging to relate biogeochemical dynamics to root				
Research area:						
This proposal is a new proposal						
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Experimental team:						
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Samples:	H2O					
-	D2O					
	SiO2 sand with traces of CaCO3 withing SiO2 container					
Instrument	t		Requested days	Allocated days	From	То
D50 T			4	4	14/09/2020	18/09/2020
Abstract:						

Experimental report for experiment UGA-111: "Combining neutron laminography with optical fluorescence imaging to relate biogeochemical dynamics to root system architecture"

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<u>Note</u>: Because of the pandemic restrictions in 2020, the experiment was slightly adapted versus what was requested in the beam time proposal. We conducted optical fluorescence imaging at University of Potsdam and derived the water content of the samples only gravimetrically instead of applying neutron radiography as planned. The samples were then sent to ILL for neutron computed laminography.

Scientific background

Respiration by plant roots and microbes is one of the most extensive CO₂ fluxes in cropped soils (Hanson et al. 2000). Gradients of O₂ and CO₂ form around roots and their extent and magnitude strongly depends on local soil water content. Localizing roots and their distribution in soil along with the observation of root respiration is achieved by the combination of optical fluorescence imaging with planar optodes (sensor foils sensitive to e.g. CO₂ or O₂) with neutron computed laminography (NCL) (**Fig. 1**). Flat, laterally extended rhizotrons allow for the measurement of biogeochemical gradients on larger scales and are optimal for planar optode imaging. NCL is a specific tomographic approach designed for the 3D investigation of slab-shaped objects. We previously successfully tested the combination of NCL and planar optodes sensitive to pH and O₂ (Rudolph-Mohr et al. 2021). CO₂ optodes we not yet combined with neutron imaging, so this combination is new and unique. In experiment UGA-111, we performed CO_2 / O_2 optode imaging (at University of Potsdam) together with 3D NCL (at NeXT Grenoble) to

a) Study root respiration related to root function, size and relative position to the optode by combined imaging of 2D CO_2 and O_2 distribution and 3D root system architecture and

b) Obtain an extended dataset that can be used to set-up and calibrate a rhizosphere model



Fig. 1 Information derived from neutron computed laminography (left) and fluorescence imaging with optodes (right). The co-registration is currently in progress.

Sample preparation, imaging experiment and preliminary results

We prepared 12 slab-shaped, boron-free glass rhizotrons ($150 * 150 * 15 \text{ mm}^3$) and glued each one planar optode sensitive to O₂ and CO₂ to their opposite inner sides. The rhizotrons were filled with sandy soil sieved to < 2 mm. Six germinated seeds of each white lupine (*Lupinus albus*) and maize (*Zea mays*) were planted and watered with a nutrient solution. Soil moisture was kept at 0.25 cm³ cm⁻³ until day 9 after planting. Between day 9 and 12 after planting, several drying-rewetting cycles were conducted to obtain time series of O₂ and CO₂ distribution via optical fluorescence imaging. The actual soil water content was calculated after weighing the samples prior to each image. After fluorescence imaging was finished, 9 living plants were sent to ILL. The average soil moisture content of the samples ranged between 0.1 cm³ cm⁻³ and 0.17 cm³ cm⁻³. The plants were stored in the dark after arrival to avoid a change of soil water content via transpiration. Neutron computed laminography (NCL) was then performed on each sample at a similar water content present at the end of the fluorescence image time-series. After the first set of NCL, the plants were kept in a plant growth chamber located next to the neutron imaging instrument, where similar growth conditions were present as at University of Potsdam. Soil moisture content dropped to $0.06 - 0.1 \text{ cm}^3$ via root water uptake and a second NCL was performed on each sample.

All datasets were reconstructed at ILL using internal software. Segmentation of the root systems is still in progress and is conducted in VG Studio Max 3.1 using a region-growing algorithm. Registration of laminographic data and CO_2 / O_2 distribution in the soil will be achieved in ImageJ after root segmentation is finished. One preliminary example of a reconstructed root system and the corresponding CO_2 distribution is shown in **Fig. 2**.



Fig. 2 Root system architecture and CO_2 concentration in the rhizosphere of maize sample 2. All data and analysis are preliminary. **A** Segmentation of the root system of maize sample 2 captured by neutron computed laminography. Optode position is indicated in red. **B** Timeseries of pCO₂ in the rhizosphere of maize sample 2 captured by fluorescence imaging. The rhizotrons were rewetted from the bottom to saturation and respiration activity (CO₂ release by roots) was observed.

References

- Hanson, P. J., N. T. Edwards, C. T. Garrten, and J. A. Andrews. 2000. "Separating Root and Soil Microbial Contributions to Soil Respiration : A Review of Methods and Observations P. J. Hanson; N. T. Edwards; C. T. Garten; J. A. Andrews." *Biogeochemistry* 48 (1): 115–46. https://doi.org/10.1023/A:1006244819642.
- Rudolph-Mohr, Nicole, Sarah Bereswill, Christian Tötzke, Nikolay Kardjilov, and Sascha E Oswald. 2021. "Neutron Computed Laminography Yields 3D Root System Architecture and Complements Investigations of Spatiotemporal Rhizosphere Patterns." *Plant and Soil*, no. 0123456789 (September). https://doi.org/10.1007/s11104-021-05120-7.