

# Annual Science Days 2023

# BOOK OF ABSTRACTS Block C Session C1

Carbon sequestration, roots and

amendments

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#### Session Description

#### Involved projects: MIXROOT-C, MaxRoot C

**Conveners:** Rebecca Hood-Nowotny (BIOS-BOKU), Isabelle Bertrand (INRAE), Anna Wawra (BIOS-AGES)

To reduce the effect of climate change carbon sequestration and or the implementation of negative emission technologies are essential. Sequestering carbon in soils through increased belowground sequestration, specifically increasing root carbon inputs from cropping systems could play a major role in reaching the 4 per mil targets. The most viable yet to date neglected option is through increased and deeper root production of both main and cover crops in both extensive and intensive cropping systems. In MIX and MaxRoot-C we are developing assessment methods to estimate root C inputs of both staple and novel crops in cropland, grassland and agroforestry systems across Europe. In this session we seek contributions which cover topics such as: measuring root traits, root biomass, root stoichiometry, root architecture, isotope labelling and rhizodeposition, in conjunction with Eenvironmental- factors, such as soil type, strength and fertility, to predict the effect of root systems on SOC stocks. We would also like to see results from the ongoing projects that might be of interest to the root community and the initial data or approaches from those working on modelling. In this session we hope to go beyond current knowledge, to evaluate the potential impact of promising C sequestering management interventions, such as: cover cropping, targetedbreeding, and soil management in these diverse agricultural production systems aiming at widespread adoption of more sustainable carbon sequestering and soil restorative practices.

Abstracts of Oral Presentations

Environmental conditions are ten times more important than wheat variety for

#### arbuscular mycorrhizal fungi

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Arbuscular mycorrhizal fungi (AMF) are an important component of the soil biota in most agroecosystems. The mutualistic symbiosis of plant roots and AMF allows plants to explore larger volumes of soil to acquire more water and nutrients and plays an outstanding role in root carbon allocation to soil. Wheat is one of the most important cereal crops worldwide. It is, however, not well understood, to what extent different wheat varieties associate with AMF and how this relationship is affected by environmental conditions. This knowledge could offer potential pathways towards a more sustainable use of water and nutrient resources in wheat production and increased soil C allocation. Within the EJP Soil project MaxRoot-C and in close cooperation with the H2020 project INVITE, we (i) compared the abundance of mycorrhizal structures in winter wheat roots between different varieties, (ii) evaluated the relative importance of variety and environment for the variability in root colonization by AMF, and (iii) tested the relation between mycorrhizal abundance and grain yield.

The study was conducted in the field season of 2021/2022 on ten modern winter wheat (*Triticum Aestivum*) varieties at four different European sites: Dotnuva LT, Eschikon CH, Freising DE, and Lleida ES, in three replicates each. The ten varieties had a high commercial relevance for the partner countries and differed strongly in yield based on experiments in the previous year. Roots were taken at the crop flowering (BBCH 63–65) stage from 0–15 cm soil depth, washed, stained and assessed for mycorrhizal root colonization by microscopy (McGonigle et al., 1990). We analyzed the data using generalized linear mixed models for binomial data and variance decomposition (LMG scores; Lindeman et al., 1980).

Arbuscular, hyphal, and vesicular abundance ranged from 10-59%, 20-91%, and 0-3%, respectively, across all samples. Averaged across sites, the varieties varied by 8% and 18% in arbuscular and hyphal abundance, respectively. Three varieties each at the lower and upper end of the abundance ranges differed significantly from other varieties, whereas four varieties were intermediate. The LMG scores for site and variety, respectively, were 5 and 51% for arbuscular

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abundance and 7 and 69% for hyphal abundance, indicating that site was 10 times more important for AMF than variety. In the next steps, we will further look into the site: variety interaction and test the relation between mycorrhizal abundance and grain yield.

Our findings show for the first time the response of mycorrhizal structures to different crop varieties and environmental conditions in arable settings across Europe.

Keywords: wheat genotype, arbuscules, vesicles, hyphae, environmental factors.

McGonigle, T. P., Miller, M. H., Evans, D. G., Fairchild, G. L., Swan, J. A. (1990). A new method which gives an objective–measure of colonization of roots by vesiculas arbuscular mycorrhizal fungi. New phytologist, 115: 495-501.

Lindeman, R. H., Merenda, P. F., Gold, R. Z. (1980). Introduction to Bivariate and Multivariate Analysis. Scott, Foresman, Glenview, IL

#### Soil intrinsic limits for carbon sequestration due to C saturation

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Carbon (C) sequestration in soils has been discussed as important climate mitigation option with the potential to generate negative emissions. Agriculture requires such negative emissions since some of their greenhouse gas emissions are unavoidable and require compensation to achieve net zero. Expectation of soils contribution to climate mitigation need to come down from theoretical assumptions to realistic estimates. In order to do so the limitations for soil C sequestration need to be analysed and discussed. Here we present case studies looking at limitations that are intrinsic due to the soils' ability to stabilize SOC on mineral surfaces (C saturation) and the current state of knowledge. More root C input for soil C sequestration maybe hampered by soils reaching C saturation. For the start of this analysis we used data of the first German Agricultural Soil Inventory comprising more than 3000 sites and fractionated a subset into particulate organic carbon (POC) and mineral associated organic carbon (MAOC). The dataset covered a range up to 12% Corg but we could not detect a limitation of soils to stabilise C as MAOC. Moreover, we investigated three long-term field experiments with different additions of organic amendments. We found a linear relation between C input to the soil and soil carbon stocks. The sand sized fraction reacted most to C additions and the silt and clay fraction showed significant increases with C addition in two experiments, likely due to the slow turnover in this fraction. C saturation was frequently discussed as reason for preventing further built up of stabilised SOC in C-rich soils. However, based on data from long-term field experiments and the national soil inventory we challenge the perception that C saturation is a limiting factor for soil C sequestration in our soils but consider available biomass as the key limiting factor for large scale C sequestration. Thus, more root-C input can increase soil carbon no matter what the soil's initial C status is.

Keywords: Carbon sequestration; C addition to soils, agricultural management

# The root shoot database. What can the available literature tell us about the effect of management on the root shoot ratio (RSR)

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The contribution of plant roots to persistent carbon stores in soil is significant. Understanding how it might be possible to optimise root biomass production through species choice and crop management is therefore an important step towards identifying agricultural policies that increase soil carbon inputs. The root shoot ratio (RSR) is the ratio between the above and below ground biomass and is used as an index for the plant biomass allocation. The RSR is a dynamic ratio influenced by biotic factors, such as plant age and species as well as abiotic factors such as climate, soil texture, nutrient, and water availability. The Root Shoot database was developed within EJP soil project Carboseq to gather all the available data on RSRs from literature and direct communication with data holders. The goal of this data set was to identify allometric relationships that might be used to improve models of soil carbon input via crop roots as a tool for policy makers. This data set was also of value to the Maxroot project where the goal is to understand specific management practices that may result in larger RSRs thereby maximising belowground inputs. To this end we present some initial results from our time working with and developing the database. We discuss the extent to which the available data can be used to increase our understanding of this critical theme as well as approach some of the limitations of the data.

Keywords: Root shoot ratio; Soil carbon, Crop management, Maxroot, Carboseq

#### Root system architecture traits of winter wheat in a genotype x environment

#### network across a European pedoclimatic gradient

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Roots are a main source of organic carbon in agricultural soils and root system architecture (RSA) traits substantially govern water and nutrient acquisition from soils. Hence, selecting varieties of main crops with improved root system architecture has been proposed as both climate change mitigation and adaptation measures in arable farming. Up to date, only little is known about the variability in root parameters of modern, commercially relevant crop varieties. Moreover, there is a lack of data on the impact of different pedoclimatic conditions across Europe on root parameters that would allow active root and, with this, soil carbon management in agriculture via variety selection.

Within the EJP Soil project MaxRoot-C and in close cooperation with H2020 INVITE, we compared a set of 10 modern winter wheat varieties for their belowground performance on 11 sites with three replicates per site. The sites spanned the major European bioclimatic regions from the Mediterranean to the Nemoral and from the Atlantic to the Pannonian zone. In the present study, we will present results of seven of the 11 sites. We sampled roots after harvest by collecting all crown roots from 0.25 x 0.25 x 0.15 m soil monoliths and by taking soil cores to 1 m soil depth. The roots were thoroughly washed, weighted and scanned and seven RSA traits (*total length, surface area, volume, diameter, orientation, length density, number of tips*) were measured using the image analysis software RhizoVision Explorer (Seethepalli et al., 2021). Additionally, the 10 winter wheat varieties were genotyped using a 25k SNP chip to further compare the relatedness of the varieties and describe genome x environment interactions in root traits. We tested differences in RSA traits between varieties using simple linear and linear mixed effects models and performed a heritability analysis on the same RSA traits.

The variability in RSA traits of the crown roots was 2.4 to 3.2 times greater among sites than among wheat varieties and all studied RSA traits differed significantly between sites. While only root surface area of the crown roots differed significantly between varieties, we found significant interactions between site and variety for the majority of RSA traits. Based on the preliminary analysis of the genetic relatedness, two out of the 10 wheat varieties were considerably different from the other eight and also from each other. In addition, we will present RSA traits of the root samples retrieved from the soil cores, with a particular focus on deep roots, and results from integrated data analyses of genome and root data.

Our preliminary findings on the crown roots imply that modern, commercially relevant wheat varieties show little variability in RSA traits and that breeding programs opting for a change in root parameters might need to consider introgression of historical accessions. However, all varieties showed substantial plasticity in RSA traits depending on the environmental conditions, indicating that root management needs to take pedoclimatic boundaries into account in order to increase carbon inputs to agricultural soils.

Keywords: root system architecture, wheat, genome x environment, deep soil, variety choice

Seethepalli, A., Dhakal, K., Griffiths, M., Guo, H., Freschet, G. T., & York, L. M. (2021). RhizoVision Explorer: open-source software for root image analysis and measurement standardization. *AoB Plants*, *13*(6), plab056. <u>https://doi.org/10.1093/aobpla/plab056</u> Increasing root-derived soil carbon input to agricultural soils by genotype selection

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Climate change mitigation and adaptation is a major challenge of modern agriculture. In general, atmospheric carbon (C) can be taken up by plants via photosynthesis and incorporated into their biomass. When the plant gets harvested, the remaining above and below-ground biomass supplies the soil with organic C. The increased incorporation of atmospheric C into soils is a promising agricultural management tool for mitigating climate change. In order to build up soil organic carbon stocks in agricultural soils or even maintaining them under the pressure of climate change, increased organic C inputs are needed. In agricultural soils, crop roots are the major source of organic C and of great importance for long-term C storage in soils as their turnover is 2 to 3 times slower than that of above ground biomass. This suggests, that genotype selection towards increased root biomass may enhance root C inputs and could therefore be a promising, easy-to-implement agricultural management option for increasing C stocks and possibly allow for additional C sequestration.

We compiled data from 13 global studies with field experiments in order to estimate the potential of optimized genotype selection to enhance root biomass without compromising yield for winter wheat, spring wheat, silage maize, winter rapeseed and sunflower. A median root C increase of 6.7 % for spring wheat, 6.8 % for winter rapeseed, 12.2% for silage maize, 21.6 % for winter wheat and 26.4 % for sunflower would be possible without yield reduction. This approach suggested a genotypic variation of root biomass but could not depict whether biomass allocation is also affected by genotype x environment interaction. To quantify this variation on the example of winter wheat, we assessed root biomass, vertical root distribution to 1 m soil depth and root: shoot ratios in a set of 10 different genotypes grown at 11 experimental sites, covering a large European climatic gradient. Preliminary results show a broad intra-specific variation in biomass production and its allocation between roots and shoots amongst the varieties. It seems possible to simultaneously select genotypes with higher grain yield and higher root biomass production meeting the needs for both food production and increased SOC build up. Additionally, increased root biomass due to deeper roots may stabilise yields under future climate change conditions where increased frequency of drought

events during vegetation periods are expected and may therefore be a climate change adaptation measure that increases the crops resilience towards changing environmental conditions.

**Keywords:** root biomass, root carbon inputs, root to shoot ratio, climate change mitigation, carbon sequestration

#### Abstracts of Poster Presentations

#### Carbon sequestration potential of legume-cereal intercropping

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Under Danish conditions, grain legumes play a crucial role in enhancing soil carbon (C) sequestration and nutrient cycling. Therefore, optimization of cultivation of grain legumes is highly important. Intercropping is believed to provide higher environmental benefits compared to monocrops. The benefits include a wide range of ecosystem services, such as a more diverse array of plant species in the field which can provide root interactions, greater above and below ground biomass, and thus increased carbon storage. However, there is a lack of comprehensive studies on the intercropping effect on legume-cereal C inputs via phyllo- and rhizodeposition (ClvPR). A field experiment with <sup>13</sup>CO<sub>2</sub> enriched atmosphere labeling-cylinders was conducted with pea, spring barley and intercropped pea and spring barley to investigate how plant C inputs into the soil in deep layers (1 m) are affected in relation to the selected species monocrop x intercropping. Results will include how the selected species differ in C allocation, and how grain legume contribute to the quantity of C input into the soil through phyllo- and rhizodeposition (qClvPR) compared to spring barley monocrop. We expect complementary effects of intercropped pea and spring barley to increase C inputs, due to better resource utilization and increased biomass in relation to plant diversity. In addition, it is expected that intercropping will have greater rooting depth soil exploration in terms of root diversity. We will report the aboveground and belowground biomass of legume and cereal monocrop which are expected to have lower densities when compared to intercropping, due to higher diversity of intercropping species allowing for the presence of a more diverse range of plant interactions.

Keywords: Crop diversification; Phyllodeposition; Rhizodeposition; leguminous

# Main crop effect on biodiversity expression in spontaneous flora and C input from cover crop mixtures

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Cover crop (CC) mixtures are believed to provide a better and wider range of ecosystem services in agroecosystems, compared to CC in pure stand. These services include increased biodiversity through increased plant diversity and carbon (C) storage through cover crop biomass and phyllo- and rhizodeposition. However, little to no information exists about how the C storage capabilities of CC mixtures are affected by the previous crops in the rotation, or how CC mixtures interact with biotic factors, such as spontaneous flora (SF), in an agroecosystem. A field experiment using isotopic labelling with "CO<sub>2</sub> was conducted to examine the impact of two different types of CC treatments (pure stand versus mixture) and preceding main crops (Pea, Barley, Pea: Barley, and Faba bean) on the input of C into the soil to 1 meter depth, with the goal of determining whether the use of diverse crops in time and space could enhance the storage of soil C in agroecosystems. Additionally, the relationship between diversified CC and SF diversity was investigated in treatments without CC, and with CC in pure or mixed stands. Results will be presented on how mixed CC contribute to the quantity of C input to the soil through phyllo- and rhizodeposition (qClvPR) and biomass compared to CC in pure stand. We expect complementary effects of different CC species in mixed stand to increase C inputs, due to better resource utilization and increased biomass. Due to complementarity, mixed CC are also expected to have higher total biomass production and greater rooting depths. Generally, CC grown after a leguminous main crop, are expected to have higher aboveground and belowground biomass. A deeper root growth is also expected, due to the N<sub>2</sub> fixing abilities of legumes which increase nutrient availability for CC growth. Cover crops following a leguminous main crop are expected to have lower relative net C inputs to the soil via phylloand rhizodeposition (%ClvPR) compared to CC grown after a non-leguminous crop, due to higher microbial activity in the fertile soil of leguminous crops. We will report how mixed CC affects densities of SF compared to no CC or CC in pure stand, where we expect mixed CC species to better capture resources and thereby suppressing SF, but at the same time supporting a higher

diversity of SF compared to CC in pure stand as they decrease the dominance of a few SF species,

allowing for the presence of a more diverse range of species.

**Keywords:** Mixed cover crops, phyllo- and rhizodeposition, carbon storage, biodiversity, spontaneous flora

#### In situ "C isotope labelling of winter wheat to determine net belowground carbon

#### inputs

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Promoting cropping systems with higher soil carbon sequestration is indispensable to reduce the effect of climate change. Plant photosynthesis and carbon allocation belowground is the primary pathway for C to enter the soil. However, little is still known about the distribution of roots and the amount of rhizodeposits for different crop varieties. Selecting varieties with increased belowground plant carbon (root and rhizodeposition) can be a viable option to maximize carbon input to the soil to enhance soil organic carbon (SOC). This study aims to determine the carbon allocation from shoots to roots and the carbon loss from these two pools to the soil in four winter wheat (WW) varieties under field and different pedoclimatic conditions in Europe.

In a replicated pan European experiment, cylinders were inserted at a depth of 25 cm before the tillering stage to confine the root system of the selected WW varieties and to control the spread of the <sup>13</sup>C tracer. Currently, we are labelling the selected varieties by <sup>13</sup>C multiple pulse labeling throughout the active growth period. After ripening, aboveground biomass will be harvested, the cylinders will be excavated, and soil samples will be taken at a depth of 1 meter. The carbon content and the <sup>13</sup>C in soil, roots, aboveground biomass, and microbial pools will be assessed by Elemental Analyzer Isotope Ratio Mass Spectrometer (EA-IRMS) and will allow us to determine net rhizodeposition. Furthermore, we are interested in determining the contribution of belowground carbon inputs to SOM by studying its molecular structure as well as in investigating the recalcitrance of this inputs through SOM fractionation.