Trace metal metabolism in plants

PLANTMETALS

annual meeting

- Co-located with, and scientific symposium integrated into, ICOBTE/ICHMET 2023

CA19116 Plant Metals

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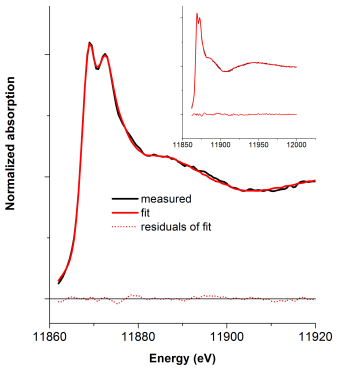
Location: Bergische Universität Wuppertal, Germany



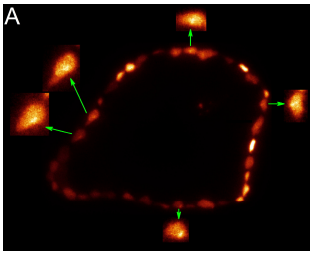
Programme:

5 September

15:00 to 17:00: Registration

17:00 to 19:00 Opening, STSM session

19:30-24:00: "Get together"- party

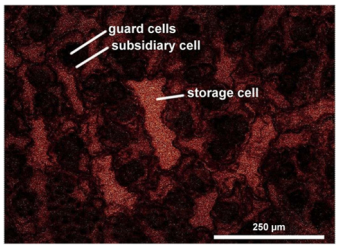


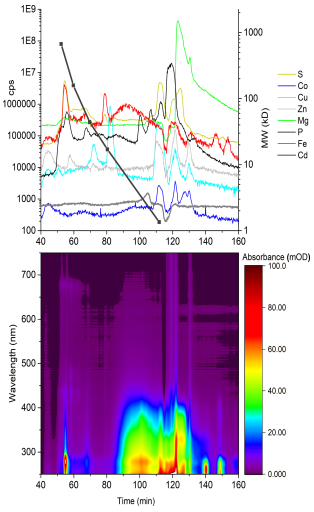
6 September

8:30-12:00 Workgroup planning

12:00-14:00: Lunch break

14:00-17:30: Management committee meeting (MC4)





7 + 8 September

8:30-18:30: Scientific symposium integrated as “special session” into ICOBTE / ICHMET

https://icobte-ichmet-2023.com

9 + 10 September (only for those registered to ICOBTE/ICHMET)

Continuation of ICOBTE/ICHMET incl. excursions

cost EUROPEAN COOPERATION IN SCIENCE & TECHNOLOGY

Funded by the European Union

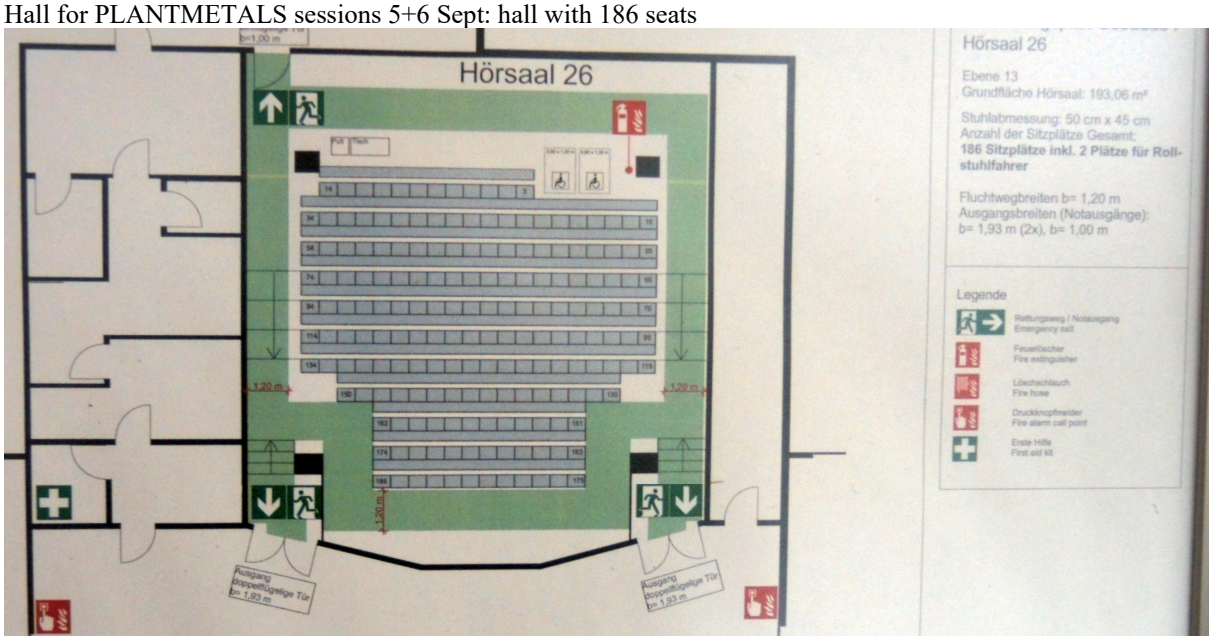
Metals, Plants and People

Detailed schedule for the PLANTMETALS meeting co-located with ICOBTE/ICHMET, state 17/08/2023

5 Sept:
Registration, welcome, STSM session

Time (start)	PLANTMETALS activity	other ICOBTE/ICHMET activity
15:00	Registration, poster mounting	
15:30		
16:00		
16:30		
17:00-19:00	- Opening / Welcome by the PLANTMETALS chair, followed by - STSM presentations (chaired by Elisa Andresen & Robert Dulfer): Up to 6 latest completed STSM's of members who are present in the meeting, 15 min each	
19:30- 24:00	PLANTMETALS "get together" party in Restaurant "Atelier", 30 € / person	

Hall for PLANTMETALS sessions 5+6 Sept: hall with 186 seats



6 Sept:
WG and MC planning

Time (start)	**PLANTMETALS activity**	**other ICOBTE/ICHMET activity** ONLY with full ICOBTE registration
8:00	-	
8:30	WG activity report and planning: WG 1 Sébastien Thomine / Ana Assuncao	
8:50	WG activity report and planning: WG2 Eva Freisinger / Marie-Theres Hauser	
9:10	WG activity report and planning: WG3 Marek Vaculík / Dorina Podar	
9:30	Coffee break	
11:00	WG activity report and planning: WG 4 Valerie Bert /Giancarlo Renella	
11:20	WG activity report and planning: WG5 Seçkin Eroğlu / Alexander Lux	
11:40	WG activity report and planning: WG6 Muhammad Imran / Mark Aarts	
12:00	Lunch break, informal discussions between participants, **continued with poster session / meeting of and with young scientists for non-MC members until end of MC meeting**	
14:00	MC Meeting (MC4): agenda defined by COST plus points added by our own. (Chaired by Hendrik Küpper & Nathalie Verbruggen) 1. Welcome, verification of the quorum 2. Information to the MC a) Recap of last meeting, e-votes b) Core Croup: report from the Core Group c, d) Action and WG Membership development e) Budget status: summary from Grant Holder. f) Update from the COST Association 3. Follow up and discussion on the a,b) Action management, COST Policies c) Grant Awarding by the Action d) WG Progress evaluation e) Science Communication Plan --> Seçkin Eroğlu f) Progress on MoU Objectives, deliverables,...	
15:30	Coffee break	
16:00	MC meeting continued : agenda defined by COST: 4. Planning of current and next WBP, incl. timing and locations of meetings and training schools 5. Monitoring and Reporting to COST: PR2 6. “AOB”: all other tasks, input from MC 7. Summary of MC decisions	Registration
17:30 -21:00		Welcome reception – “icebreaker” – included in ICOBTE registration fee – best chance to start networking with ICOBTE attendants who are not PLANTMETALS members

7 Sept: PLANTMETALS Scientific Symposium part 1
Oral talks selected by the WG leaders, plus plenary oral talk by ICOBTE-invited PLANTMETALS vice chair Nathalie Verbruggen, Posters

Time (start)	**PLANTMETALS activity**	**other ICOBTE/ICHMET activity** ONLY with full ICOBTE registration
8:00	-	Registration
8:30	Poster session 1 (Those fully registered to ICOBTE use the first half hour for a quick look and attend the opening ceremony afterwards)	
9:00		Opening Ceremony hall with 788 seats
9:30		
10:00		
10:30	coffee break	
11:00	for those with full ICOBTE registrations: plenary talks, others: networking / posters	Plenary talk 1: Tanja Schwerdtle
11:45		Plenary talk 2: Donald Sparks
12:30	lunch	
13:00		
13:30		
14:00-15:30	Oral session 1, (chaired by Marie-Theres Hauser & Nathalie Verbruggen)): 1 keynote talk (20min + 5min discussion), Ana Assuncao, 14:00-14:25 4 talks selected from abstracts (15 min): Claudia Cosio 14:25-14:40 Zheng HongXiang (14:40-14:55 Hendrik Küpper (14:55-15:10) Edgar Peiter (15:10-15:25) <5 min spare time for discussion>	Oral session
15:30	coffee break	
16:00-17:30	Oral session 2 (chaired by Marek Vaculik & Elisa Andresen): “poster talks” selected from abstracts (8 min – discussion ONLY in the poster session at the posters!) afterwards: group photo Emre Aksoy Anna Barabasz Adriana Mišúthová Eva Freisinger Filis Morina Nadege Oustriere Diego Baragano Abdel Rahman Al Tawaha Maria J. Poblaciones Matthias Wiggenhauser (presented by Josip Jurkovic) Hiram Castillo-Michel	Oral session
17:30-18:30	Joined PLANTMETALS-ICOBTE poster session A	
18:30-19:00		
19:30-24:00		Beer and Dinner in Brewery

Hall for plenary talks
– please note that plenary talks can ONLY be attended by those who fully registered for ICOBTE!

8 Sept
Oral talks selected by the WG leaders, plus plenary oral talk by ICOBTE-invited PLANTMETALS vice chair Nathalie Verbruggen

Time (start)	PLANTMETALS activity	other ICOBTE/ICHMET activity: ONLY with full ICOBTE registration
8:15	Oral session 3 (chaired by Hendrik Küpper & Filis Morina) 1 keynote talk (Anthony van der Ent, 8:15-8:40), <20min +5 min discussion> 5 regular talks selected from abstracts, 15 min incl. discussion: Giancarlo Renella (8:40-8:55) Oskar Siemianowski (8:55-9:10) Valérie Bert (9:10-9:25) Aida Bani (9:25-9:40) Michel Mench (9:40-9:55) <5 min free for discussion>	Oral session
10:00	coffee break	
10:30	Poster session 3	Oral session
12:00	lunch	
13:30		**Plenary talk: Nathalie Verbruggen**
14:15	Poster session 4	Plenary talk: Mark Saito
15:00	coffee break	
15:30	Poster session B of PLANTMETALS	Oral session
17:00	Joined poster session B of PLANTMETALS and ICOBTE/ICHMET	
17:30		
18:00	poster de-mounting, free evening	
18:30-19:00		
20:00-24:00		Gala dinner

Poster area of PLANTMETALS

9 September: ICOBTE programme, only for members with full ICOBTE registration

- The full programme can be found at https://icobte-ichmet-2023.com/
- **Including plenary talk by ICOBTE-invited PLANTMETALS member Fangjie Zhao**

Time (start)	other ICOBTE/ICHMET activity: ONLY with full ICOBTE registration
8:30	Oral session, incl. Regular session 7: „Plants and trace elements" (same lecture hall as previously PLANTMETALS lectures)
10:00	coffee break
10:30	Oral session, incl. Regular session 7: „Plants and trace elements" (same lecture hall as previously PLANTMETALS lectures)
12:00	lunch
13:30	**Plenary talk: Fangjie Zhao**
14:15	Plenary talk: Jurate Kumpiene
15:00	coffee break
15:30	Oral session, incl. Regular session 7: „Plants and trace elements" (same lecture hall as previously PLANTMETALS lectures)
17:00-18:30	Closing Ceremony

10 September: ICOBTE excursions, only for members with full ICOBTE registration

AKBUDAK, M. Aydin

PdTMT: A Novel Thiocyanate Methyltransferase (TMT) from European Alkali Grass (Puccinellia distans) Involved in Selenium Tolerance

KUBRA BUDAK; M.AYDIN AKBUDAK
Akdeniz University - Turkey (TR)

Selenium is a metalloid that is found naturally in the environment, including in soil, water and air. While it is essential for proper plant growth and development, excess selenium can lead to toxicity in many plant species. However, some wild plant species, such as Astragalus spp. and Puccinellia spp., are more tolerant to selenium toxicity and are able to withstand higher levels of the element. In our research, we employed a yeast cDNA expression library screening method to identify genes in European Alkali Grass (Puccinellia distans) that may allow the plant to tolerate selenium toxicity. The yeast (Saccharomyces cerevisiae) transformants expressing different cDNAs were selected for their ability to survive under toxic selenium conditions (10 mM Na2SeO3). Phylogenetic analyses revealed that cDNAs isolated from resistant colonies showed approximately 90% sequence similarity with the predicted “thiocyanate methyltransferase” (TMT) genes of Triticum urartu, T. aestivum and T. dicocoides. Serial dilution experiments showed that yeast cells with PdTMT cDNAs continued to survive at Na2SeO3 concentrations up to 50 mM.

The expression of PdTMT in P. distans increased over time in response to selenium stress, reaching a maximum level of 5.27-fold (log2) after 48 hours of exposure to 80 µM Na2SeO3, which is a toxic concentration for this species. The expression of PdTMT was significantly increased by Na2SeO3 in yeast cells, reaching a maximum of 21.86-fold (log2) at a concentration of 10 mM. Thiocyanate methyltransferase (TMT) catalyzes the transfer of a methyl group from S-adenosyl methionine (SAM) to thiocyanate (SCN-) which is a toxic compound that can interfere with the function of certain enzymes and disrupt normal cellular processes. By methylating SCN-, TMT may help protect cells from the harmful effects of SCN-. This is the very first study to examine any TMT gene conferring tolerance to a metal/metaloid toxicity. It is thought that the results obtained will contribute to the development of varieties tolerant to metal/metalloid stress in breeding studies.

AKIN, Melekşen

The Role of Biochar and boron fertilizers application on growth and Productivity of Faba bean (Vicia faba L).

Melekşen Akın[1] & Sadiye Peral Eyduran[2]
[1] Department of Horticulture, Iğdır University, Iğdır 76 000, TURKEY
[2] Department of Horticulture, Muğla Sıtkı Koçman University, Fethiye, TURKEY

We performed a bibliometric analysis on ecotoxicology utilizing Bibliometrix package available in R studio. The bibliographic data consisted of 7156 documents published until 2023 in Web of Science database. Our results demonstrated an upward trajectory in the publication trend with around 16% annual growth. The data was released in 1385 sources with Environmental Toxicology and Chemistry being the leading journal on the area followed by Science of the Total Environment. The USA followed by France and Brazil had the highest number of publications. The most prominent countries on the area did not show high international collaboration but rather had single country publications. The most frequent author keywords projected after ecotoxicology were risk assessment, biomarkers, pesticides, toxicity, heavy metals, fish and bioaccumulation which represent the current direction of the research field. Other significant keywords were oxidative stress, biomonitoring, genotoxicity, cadmium, copper, zebrafish, Daphnia magna and nanoparticles. Collaboration network analysis was run to show hidden collaboration patterns on the field among countries, institutions and authors which resulted in two subnetworks among countries, eight among institutions and nine subgroups among authors.

Keywords: science mapping, thematic evaluation, network analysis

AKSOY, Emre

GATA12 regulates iron deficiency responses by binding to the promoters of BHLH transcription factors in Arabidopsis

Emre Aksoy[1]; Amir Maqbool[2]; Bayram Ali Yerlikaya[3]; Allah Bakhsh[4]
[1] Middle East Technical University (METU);
[2] Nigde Omer Halisdemir University;
[3] Ondokuz Mayis University;
[4] University of the Punjab

The lack of iron (Fe), one of the basic micro-nutrients for plants, affects plant growth and yield and restricts agricultural productivity. Although the mechanisms involved in Fe uptake from the rhizosphere and root-to-shoot translocation have been understood well, the transcription factors controlling these mechanisms were only concentrated in the bHLH family. GATA transcription factors control cell differentiation, and leaf and root development in plants. GATA9 and its close paralog GATA12 are upregulated in Arabidopsis roots under Fe deficiency. To determine the functions of GATA12 in Fe uptake and translocation, several responses of two gata12 T-DNA insertion mutants (gata12-1 and gata12-2) and two GATA12 overexpression lines (OE1 and OE2) were compared to the wild type Col-0 under Fe deficiency. Accordingly, mutant plants were more sensitive to Fe deficiency depending on the expression levels of GATA12, while GATA12 OE2 was more tolerant to Fe deficiency. This sensitivity in the roots was partially attributed to the decrease in ferric chelate reductase activity and FRO2 gene expression. While the amount of Fe accumulated in mutant roots was similar to Col-0, it was significantly decreased in the GATA12 OE lines. Following confirmation of the iron deficiency phenotype, RNA sequencing was performed to identify transcripts affected by Fe deficiency in the roots and shoots of gata12-1 and GATA12 OE plants. Bioinformatic analyses showed that GATA12 and its close paralogues GATA2, GATA4, and GATA9 formed a network together that connects the bHLH networks operating in the root epidermis and pericycle. Moreover, sulfur metabolism, and phenylpropanoid and flavonoid biosynthesis pathways were affected more by GATA12. Under Fe deficiency, 47 genes with expression levels of three and above were identified as regulated by GATA12. ChIP-RT-qPCR and EMSA analyses proved that GATA12 can bind the GATA motifs in the promoters of some of these genes, including FIT and PYE. In conclusion, it has been shown that GATA12 regulates Fe signalling above bHLH transcription factors.

AL TAWAHA, Abdel Rahman

The Role of Biochar and boron fertilizers application on growth and Productivity of Faba bean (Vicia faba L).

Abdel Rahman Al Tawaha
Department of Biological sciences, Al-Hussein bin Talal University, Maan, Jordan

A field study was conducted to investigate the role of Biochar and boron fertilizers application on Faba bean (Vicia faba L). The experimental design was factorial, with three biochar concentrations (0, 0.5, and 1 percent w/w) and four Boron treatments included the foliar application of four rates of boron 0, 1.0, 2.0, and 3.0 Kg B ha-1 applied at the vegetative stage. The variables that were measured were seed yield per plant, number of pods per plant, number of seeds per pod, plant height, pod length, number of branches per plant, number of leaves per plant, seed oil percentage, seed oil yield/plant, seed nitrogen, seed total carbohydrates, Potassium (K) percent, Phosphorus (P) percent, and Iron (Fe) percent. According to the findings, Faba bean responded significantly better with biochar application in terms of yield and related traits, as well as mineral accumulation, in comparison to plants grown without biochar application. On the other hand, a high rate of Boron Fertilization improves seed yield per plant, number of pods per plant, and number of seeds per pod. Overall, the combination of Biochar application and Boron Fertilization can improve the growth and Productivity of faba bean plant.

ALIRZAYEVA, Esmira

Root architecture of Artemisia plants under heavy metal and petroleum contamination

Esmira ALIRZAYEVA[1], Gunter NEUMANN[2], Volker ROEMHELD[2]

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Keywords: Pollution, Artemisia plants, root growth, multitolerance

Plant roots are the key component to improve resources acquisition under competitive nutritional status and tolerant plants develop the root foraging strategy through the morphological alteration. Root architecture of different Artemisia L. species and ecotypes from heavy metal (HM) and petroleum (PH) polluted areas as well as non-polluted area was comparatively studied on HM and PH contaminated soils. Although no significant difference was detected on root parameters of all species independence of origin and levels of HM contamination of soils and all plants tested demonstrated tolerance to all pollutants, some differences were observed in the root growth strategies of plants on soils with HM and PH pollution. In ecotype from PH polluted and non-polluted areas, root growth parameters were lower, but the average diameter (AD) was higher than in ecotypes from HM polluted areas when grown on the HM contaminated soils. Besides, all tested plants demonstrated similar accumulation capacity of HM, regardless of their type, origin and level. While the acceptable level of concentration of Zn and Cu were found in shoots, Cd concentration reached the toxic level. On PH contamination soils, the increase in number and decrease in length of root hairs in the ecotype from PH polluted area was observed. Non-adapted ecotype displayed higher development under HM than PH contamination with higher AD and less length and number of lateral roots. Plants from PH polluted area were found to develop thicker roots, consequently lower specific root length, also higher volume and surface area than the ones from HM contaminated and non-contaminated areas in their contrasted original habitats. This development traits can be considered as better establishment and penetration capacity of the roots for the exploration of the soil profile under imbalanced nutritional conditions.

ANDRESEN, Elisa

Low level Cd toxicity to soybeans affects the root metabolome, cellular distribution of Cd and reveals high affinity protein binding sites

Elisa Andresen[1]; Isvett Josefina Flores-Sanchez[1]; Dennis Brückner[2]; Syed Nadeem Hussain Bokhari[1]; Gerald Falkenberg[2]; Hendrik Küpper[1]

[1] Czech Academy of Sciences, Biology Centre, Institute of Plant Molecular Biology, Department of Plant Biophysics and Biochemistry, Branišovská 31/1160, 370 05 České Budějovice, Czech Republic
[2] Deutsches Elektronen-Synchrotron DESY, Notkestr. 85, 22607 Hamburg, Germany
[3] University of South Bohemia, Faculty of Science, Department of Experimental Plant Biology, Branišovská 1760, 370 05 České Budějovice, Czech Republic

Even low concentrations of Cd can be unfavourable for plants, inducing chronic toxicity, reducing biomass and yield and potentially enter the human food chain. We analysed non-toxic (0.5-50 nM), sublethally toxic (< 550 nM) and lethally toxic (3 µM) Cd concentrations in soybean plants (Andresen et al., 2020; 2023). Here we show the concentration dependent changes in the mechanism of Cd toxicity in soybean roots in terms of tissue-level and subcellular Cd localisation and Cd binding to proteins. Cd deposition was predominantly inside cortex cells (likely vacuoles) at sublethal concentrations, but in cell walls of the outer cortex and the vascular bundle at lethal concentrations, as determined by synchrotron µXRF-CT measurements. The Cd binding to proteins drastically changed too, with different proteins being the main targets at sublethal vs. lethal Cd toxicity. The likely high-affinity binding protein targets in roots were identified by protein purification from natural abundances. They were allantoinase and protein disulphide isomerases at low Cd, but aquaporins, peroxidases and cupredoxins dominated at higher Cd concentrations. This differed from previous results in leaves, where the main target was clearly the light harvesting protein LHCII (Andresen et al., 2020). A vast change in several metabolites and lipophilic compounds indicated induction of flavonoids and phytoalexins as well as membrane-reorganisation.

References:

Andresen E, Flores-Sanchez IJ, Brückner D, Bokhari SNH, Falkenberg G, Küpper H. 2023. Sublethal and lethal Cd toxicity in soybean roots specifically affects the metabolome, Cd binding to proteins and cellular distribution of Cd. J. Haz. Mat. 442: 130062

Andresen, E, Lyubenova, L, Hubaček, T., Bokhari, S.N.H., Matoušková, Š, Mijovilovich, A, Rohovec, J, Küpper, H, 2020. Chronic exposure of soybean plants at nanomolar cadmium reveals specific additional high-affinity targets of cadmium toxicity. J. Exp. Bot. 71, 1628–1644

Funding: COST (CA19116 “Trace metal metabolism in plants – PLANTMETALS), Ministry of Education, Youth and Sports of the Czech Republic with co-financing from the EU (KOROLID, CZ.02.1.01/0.0/0.0/15_003/0000336), Czech Academy of Sciences (RVO: 60077344).

ASSUNÇÃO, Ana

Dealing with too little: The Zn Deficiency Response in land plants

Ana Assunção
University of Copenhagen - Denmark

All organisms require zinc (Zn) as an essential micronutrient due to its structural and catalytic role in many proteins. Membrane transporters and low-molecular-weight ligands play a major role in maintaining an adequate Zn distribution and intracellular availability, which requires a tight regulation to avoid Zn deficiency or toxicity.

In Arabidopsis thaliana, bZIP19 and bZIP23 transcription factors are the central transcriptional regulators of the Zn deficiency response. They are F-group basic leucine-zipper proteins (F-bZIP), characterized by a cysteine(Cys)/histidine(His)-rich motif domain. bZIP19 and bZIP23 transcription factors also function as sensors of intracellular Zn concentration, through direct binding of Zn2+ ions to the Cys/His-rich motif, now termed Zn-sensor motif (ZSM). bZIP19/23 activity regulate cellular Zn levels through Zn-dependent changes of their target genes expression. These targets encode Zrt/Irt-like protein (ZIP) transporters, involved in cellular Zn uptake, and NAS enzymes that produce the Zn ligand nicotianamine (NA), involved in Zn distribution. Interestingly, mutations in the ZSM of AtbZIP19 led to a Zn-independent activation of the target genes, and to a significant increase in Zn concentration in leaves and seeds.

There is evidence for evolutionary conservation of the F-bZIP-regulated Zn deficiency response across land pants, which opens opportunities for translational approaches to develop crops with improved Zn nutritional content (biofortified) and improved resilience to Zn-deficient soils. This is significant considering that Zinc-deficient soils are widespread globally and human Zn malnutrition affects about one-third of the world's population.

Here, I will present an overview of the research developed at my Lab, where we investigate these and other aspects of the F-bZIP regulatory network, including the F-bZIP Zn-sensor function and the role of F-bZIP targets in the Zn deficiency response. New findings include analysis of the effect of AtbZIP19 ZSM mutations on plant performance and ionome profile and discussion of its relevance and application in crops. Finally, the integration of this knowledge in an evolutionary perspective is also addressed.

AUGUSTYNOWICZ, Joanna

Application of filamentous algae and symbiotic bacteria in a model in vitro study on Cr(VI) bioremediation mechanisms

Joanna Augustynowicz, DSc[1]; Patrycja Fidelus[1]; Anna Kowalczyk[2]; Konrad Wołowski ProfTit[3]; Alina Wiszniewska, DSc[1]; Dariusz Latowski, DSc[2]

[1] University of Agriculture in Krakow;

[2] Jagiellonian University in Krakow;

[3] W. Szafer Institute of Botany, Polish Academy of Sciences

Cr(VI) has extremely toxic properties due to its very high bioavailability and reactivity. In our earlier work (Environmental Pollution 2021) we described unique "natural bioreactor" – a retention ditch contaminated with Cr(VI) 6,000 times over the limits for more than 20 years. This "bioreactor" was a habitat for filamentous algae exhibiting hyperaccumulation towards Cr, among which Stigeoclonium sp. (Chlorophyceae) was found in large quantities. The aim of the present work was to create an original model for the study on hexavalent chromium bioremediation mechanisms in a biological system made of filamentous algae and accompanying bacteria. We isolated Stigeoclonium sp. from this real-world environment and then successfully introduced into in vitro conditions. A part of the Stigeoclonium sp. culture was treated with timentin - an antibiotic used in plant biotechnology with a broad spectrum of bacteriostatic and bactericidal activity. We detected very significant decrease in the biomass growth and the drop in the photosynthetic pigment contents in the algae growing after the antibiotic treatment. Interestingly, addition of Cr(VI) into the solution did not limit the growth of algae of the both types of cultures, though it had very pronounced impact on filament's morphology stimulating their branching. Nuclei isolated from timentin-treated and untreated algal cells were used to assessed for DNA integrity and potential genotoxic effects of chromate. Removal of Cr from the solution and detoxification of Cr(VI) by its reduction to Cr(III) was also evaluated in these two types of cultures. Finally, after the isolation and characteristics of the symbiotic bacteria, the role of the particular isolates in the protection and/or stimulation of the algae during Cr(VI) bioremediation is suggested.

BALOCH, FAHEEM SHEHZAD

Germplasm characterization for micro and macronutrients in bean and sorghum and linkage disequilibrium for identification of genomic regions for future food security to eliminate hidden hunger

Faheem Shehzad Baloch1*, Muhammad Azhar Nadeem1, Mehmet Bedir1, Muhammad Tanveer Altaf1, Waqas Liaqat2, Gönül Cömertpay2
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[2]Eastern Mediterranean agricultural research institute, Adana, Türkiye
*balochfaheem13@gmail.com

The world is confronted with food insecurity due to climate change and nearly 800 million people from developing countries go to bed hungry. Characterization of genetic resources has always remained one of the favorite methods of scientific community to investigate the novel variations which can be used for the development of improved cultivars expressing higher yield with better quality, biotic and abiotic stress resistance. Bean is cultivated in nearly all parts of the world and its bean seed composition with respect of mineral and biochemical has been improved significantly during recent years. Similarly, Sorghum production is steadily increasing worldwide, especially because it has high yield potential, as well nutritional properties of sorghum grain under climate change scenario. Present investigation was done under different projects from ERANET and TÜBİTAK for investigation of genetic variation for mineral, antioxidant, protein and vitamin traits. A total of 183 bean landraces were characterized under two environments for three years and 172 sorghum diversity panel was investigated under limited conditions of H2O and N. Plentiful diversity was observed in both crops and bean and sorghum genotypes were identified with stable and high mineral elements (Zn, Fe, Mg, Mn etc) and nutritional properties. GBS analysis was performed in bean and whole genome resequencing in sorghum resulted in thousands of SNPs for genome wide association studies (GWAS). Some highly significant SNPs linked with mineral, antioxidant, vitamins and protein were identified in both crops. This huge genetic variation and genomic tools in both crops can be effectively used for genomic breeding for developing the bean and sorghum varieties with desired mineral and quality traits to feed the World population to cope with hunger hunger

Acknowledgment: The results are part of three different projects. Therefore, authors are very grateful to ERANET (220N276) and TÜBİTAK (TOVAG 215O630 and 120O920) for financial supports for conducting these researches

BALTRĖNAITĖ-GEDIENĖ, Edita

Does biochar amendment to agricultural soil cause a risk of the formation of environmentally persistent free radicals?

Edita Baltrėnaitė-Gedienė[1]; Slawomir Lomnicki Ph.D.[2]; Chuqi Guo[2]
[1] Vilnius Gediminas Technical University;
[2] Louisiana State University

Environmentally persistent free radicals (EPFRs) have been considered emerging contaminants due to their harmful effects on human health. The adverse health impacts are attributed to oxidative stress induced by EPFRs through forming of reactive oxygen species (ROS).

Biochar is widely considered a valuable organic amendment for agricultural and environmental remediation purposes. Biochar production via pyrolysis entails the thermal decomposition of organic compounds in the feedstock, with the carbonisation conditions and feedstock type assisting the formation of EPFRs. When biochar is applied on the soil, EPFRs may promote the formation of ROS, and thus influence the transformation of organic and inorganic contaminants in soil and impact the rhizosphere. Therefore, agricultural soils may become a source of EPFRs. However, the fate and transformations of EPFRs in soils after biochar amendment needs to be better understood and studied.

Different soil cultivation types, the addition of fertilisers and variation in biochar input, on the one hand, and the presence of metals in soil, biochar and fertilisers, on the other hand, will be discussed to provide different conditions for EPFRs formation, accumulation and fate in agricultural soils.

BANI, Aida

Phytoextraction of heavy metals by Odontarrhena chalcidica cultivated on agricultural soil in industrial site of Elbasan, Albania

Aida Bani[1]; Dolja Pavlova[2]; Marilda Osmani[3]
[1] Agricultural University of Tirana, Faculty of Agronomy and Environment;
[2] University of Sofia, Faculty of Biology;
[3] University of Elbasan "Aleksandër Xhuvani" Faculty of Natural Sciences

The contamination of heavy metals has attracted the world's attention due to an intensive effect on ecosystems and its toxicity to living organisms. The heavy metals exist naturally in ultramafic rocks, but human activity has increased their accumulation. Albania has a high density of abandoned or active mining sites and metal smelters. The metallurgical complex in the industrial area of Elbasan is a potential source of heavy metal pollution in this part of the country. The pollution emitted from this complex has affected agricultural soil and the water of Shkumbin River, the main watershed in this region, and has caused many problems to the microenvironment and adversely affected human health. A three-year field experiment was performed on polluted soil in ex-metallurgical industrial site in Elbasan. The main goal of this study was the assessment of the heavy metal concentration (Ni, Co, Pb, Cr) and the evaluation of the phytoextraction technology for soil remediation. The Ni hyperaccumulator species Odontarrhena chalcidica was cultivated under some agronomic practices: soil tillage, four plants per 1 m2, organic and mineral fertilization, and irrigation. The experiment showed more effective Ni phytoextraction when 50% of "vegetation soil" was added to the industrial soil instead of manure. Also the concentration of Ni in soil was high (610 mg/kg). After three years an improvement of the soil fertility for conventional agriculture (Ni stress) was established. The Ni hyperaccumulator species O. chalcidica increased the biomass production and Ni yields under agronomic practices. The soil Ni availability was 15% lower after 3 years of successive cropping. This study showed that O. chalcidica could be an useful candidate for phytoextraction technologies. We propose phytomining as nature-based solution to restore contaminated or degraded soils while producing biomass for industrial use in the city environment.

BARABASZ, Anna

In search of mechanisms underlying the redistribution of Zn in tobacco under Zn-deficiency conditions in medium

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Root/shoot Zn distribution in plants is conditioned upon metal concentration in medium. In not-Zn-hyperaccumulating plants shoot/root Zn concentation ratio is higher in plants growing in Zn-deficiency than in plants growing in optimal conditions. We showed that Zn status-dependent root/shoot distribution of Zn was related to distinct metal accumulation in root parts. In plants growing in optimal Zn conditions the highest Zn concentration was observed in middle parts, and in Zn-deficiency in apicals ones.

The aim of our research was to determine whether and how fast shoot/root Zn amount ration increase and the Zn concentration in the middle part of the roots decreases after transplanting plants to Zn-deficient conditions and what genes participate in this process.

Our experiments showed that in 5,5-weeks old tobacco cultivated hydroponically in sufficient conditions (preculture) about 40% of the accumulated Zn was detected in the shoots. The transfer of the precultured plants to the Zn-deficient medium and their additional 6-day cultivation resulted in a change in the root/shoot Zn distribution. 50% of the accumulated Zn was detected in the shoots. In plants continuing further growth in sufficient conditions, approx. 35% of the accumulated Zn was in the shoots. Both plants grown in the sufficient and Zn-deficient conditions increased their dry weight approx. 4.5 times. The analysis of the middle part of the main roots showed that the Zn concentration in these root parts constituted 30% (deficiency) or 63% (sufficiency) of the Zn concentration detected in precultured plants. These results confirm that the middle part of the main root is a reservoir of Zn, which can then be redistributed to other parts of the plants.

Searching for genes participating in the Zn redistribution from the middle part of the root, RNAseq analysis was performer. Several genes potentially involved in Zn redistribution were selected.
On the conference obtained results will be presented.

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BARAGAÑO, Diego

Insights of cadmium isotopes in the soil-plant system to understand the remediation processes of contaminated soil using zero-valent iron nanoparticles

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Nanoscale zero-valent iron (nZVI) is effective for Cd-polluted soil remediation by reducing metal mobility and immobilizing Cd via sorption to newly-formed Fe oxides. However, the interactions between nZVI and Cd in the soil-plant system remain unknown, thus we propose the analysis of Cd isotopic composition for understanding the soil remediation process. Pot and rhizobox experiments were performed using untreated and nZVI-treated soils, with and without Agrostis capillaris, an autochthonous plant growing in the polluted site, to evaluate the effects of nZVI, plants, and their combination on Cd isotopic composition. After 45 days, plants (aerial parts in both, and also roots from rhizoboxes), soils and pore water samples were collected for measuring Cd concentration and Cd isotope ratios using a thermal ionization mass spectrometer (Triton, Thermo Fisher) based on the double spike method. The results revealed that the application of nZVI decreased the Cd concentration from 145 to 33 µg/L in pore water, and preferentially immobilized the light Cd isotopes pool (δ114/110Cd=0.20‰ to 0.55‰ respectively). Furthermore, a decrease in Cd concentration in aerial part was also detected (from 22 to 9 mg/kg) and plants prefer extracting similar pool of Cd isotopes in both cases (δ114/110Cd=0.21‰). Roots in untreated soil showed again similar Cd isotopic composition to pore water and aerial parts, although a high difference was found for the roots in the nZVI-treated soil (δ114/110Cd=-0.22‰). However, this value did not reflect Cd uptake by roots due to the presence of nZVI and newly-formed Fe oxide particles in these tissues, as was observed by electron microscope. Therefore, the isotopic composition corresponds to the Cd immobilized in these Fe-based particles, with an enrichment of heavier isotopes of Cd, consistent with the scientific literature. In conclusion, our results indicate that soil amendments such as nZVI preferentially immobilize certain pools of Cd isotopes. This highlights the usefulness of analyzing Cd isotope composition in understanding soil remediation and selecting appropriate amendments for specific plant species.

BERT, Valérie

In situ selection of crops to manage a metal-contaminated dredged sediment-derived soil: fate of metals in plants and plant effect on metal extractability in soil.

Valérie BERT; Alexandre Perlein

Ten crops, usually grown with conventional farming, horticultural or forestry practices, were chosen for their potential to be valued after harvest in non-food chains and their relevance to pedoclimatic conditions. These crops (Linum usitatissimum L., Eucalyptus sp., Beta vulgaris sub sp. vulgaris L., Phacelia tanacetifolia Benth., Malva sylvestris L., Chenopodium album L., two cultivars of Sorghum bicolor L. (Biomass 133 and Santa Fe red), and two cultivars of Salix sp. (Inger and Tordis)) were cultivated in situ on plots of a metal-contaminated dredged sediment-derived soil (DSDS) located in the North of France. The fate of metals (Cd, Zn, Cu, Pb) in these crops was studied as well as the mobility of the metals in the DSDS with or without vegetation. The measurement of the metal concentrations in the aerial plant parts and both the total and the extractable fractions of metals in the DSDS altogether with the growth pattern of the plants allowed to classify the plants in two groups, i.e. metal tolerant excluder or accumulator. Amongst the tested crops, all were metal tolerant and most exhibited an excluder behavior, which may favor their use in phytostabilization, a tool to manage polluted sites. In addition, the metal mobility in the DSDS was not increased by the plants. These results are required inputs to further assess potential environmental and heath exposure and risk, due to the presence of metals in the plants and in the DSDS, considering crop valuation after harvest. The oral presentation will be based on a synthesis of outcomes of several publications (Perlein et al. 2021a,b; 2022).

BLAUDEZ, Damien

Mycorrhizal and endophytic fungal symbionts for improving plant fitness and modulating metal accumulation in the context of the phytomanagement of metal-contaminated sites

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Plant roots establish intimate associations with a variety of fungi such endo-, ectomycorrhizal fungi and fungal endophytes. Root endophytic associations involve a diverse array of fungi, the best known are dark septate endophytes (DSEs). DSEs are a polyphyletic assemblage of ascomycetous fungi whose structure is characterized by dark melanized hyphae and microsclerotia located in plant roots. Associations with DSEs are commonly found in various biomes and plant taxa. Although DSEs are commonly recorded, the effects of their colonization on plant growth and fitness are unclear. Here, we show from lab mesocosms to large-scale field experiments that they can promote plant growth by improving nutrition (e.g. solubilization of minerals, degradation of complex carbon compounds), produce secondary metabolites (e.g. phytohormones, volatile organic compounds) and protect hosts against phytopathogens. More particularly, the high tolerance of DSEs to abiotic stresses and their relatively high abundance in trace element-contaminated and other stressful habitats suggest that they may have an important function for host survival under these conditions. The accumulation of high concentrations of melanin in hyphae is characteristic to this group and could reflect a common response to different environmental pressures. According to the different plant species they associate with, metal tolerance and/or accumulation can be modified. Finally, we outline why additional research is required in the emerging field of plant–fungus interactions to address future challenges in the context of the phytomanagement of metal-contaminated sites, especially for purposes of biomass production, agromining/phytoextraction and phytostabilization.

BLOMMAERT, Hester

The odyssey of cadmium in cacao trees

Hester Blommaert[1]; Anne-Marie Aucour; Mattthias Wiggenhauser; Umaharan Pathmanathan; Erik Edward Smolders; Géraldine Sarret
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Cadmium (Cd) concentrations in cacao often exceed food limits. The research on strategies to reduce Cd accumulation in cacao beans is currently limited by a lack of understanding of the Cd transfer pathways within the cacao tree1. We elucidated the mechanisms that control Cd transfer in a high Cd accumulating cultivar by a combination of Cd stable isotope analyses and X-ray absorption spectroscopy. The latter is providing insights on Cd speciation whilst isotopic fractionation is increasingly used to infer Cd pathways in plants. Vegetative (roots, branches, leaves) and reproductive (placenta, nib (seed), testa (seed envelope), pod husk) tissues were collected from three genetically identical trees in a conservatory of cacao cultivars in Trinidad (soil Cd: 0.3 mg Cd kg-1). The plant Cd concentrations were 10-28 higher than the topsoil Cd concentrations and increased as placenta < nib < testa < pod husk < root < leaf < branch. Mass balance estimations indicated that the largest fraction (57%) of total plant Cd was present in the branches where it was primarily bound to carboxyl-ligands (60-100%) and mainly localized in the phloem rays and phelloderm of the bark. The retention of Cd in the roots was low, and light Cd isotopes were retained in the roots while heavier Cd isotopes were transported to the shoot, as observed in annual crops. The isotope fractionation data further revealed that the main contribution of nib Cd was from the phloem tissues of the branch rather than from leaf remobilization, contrastingly to annual crops. In the nibs, Cd was mainly bound to oxygen ligands (60-90%), with phytate as the most plausible ligand. This study extended the limited knowledge on Cd accumulation in perennial, woody crops and revealed that the Cd pathways are markedly different than in annual crops, which has implications for mitigation strategies2.

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BOČAJ, Valentina

Inter- and intraspecies differences in microbiome and transcriptome of two hyperaccumulating Noccaea species from Slovenia

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Hyperaccumulators are plants that can accumulate high concentrations of one or more metal(loid)s in their above-ground biomass, especially leaves, without showing any visible toxicity symptoms (Brooks et al., 1997). Prior studies have shown that the microbiome differs between compartments (i.e., different spatial niche) and may play an important role in hyperaccumulation (Kushwaha et al., 2022; Vogel-Mikuš et al., 2006). The resistance/tolerance genes for specific metal(loid) seem to be very abundant in hyperaccumulators, and their abundance increases with the increasing concentration of the metal(loid) in the soil (Xiao et al., 2021). We still know very little about the microbiome and transcriptome in hyperaccumulators and comparisons of the microbiome and/or transcriptome of closely related hyperaccumulators grown under the same environmental conditions remain scant. Therefore, in spring 2022, two closely related hyperaccumulator species, namely Noccaea praecox and Noccaea caerulescens, were sampled from two sites in Slovenia: a non-polluted site in Lokovec where both species co-occur and a polluted site in Žerjav where only N. praecox grows (Likar et al., 2010). For metagenomic analyses, bulk and rhizosphere soil were used as well as plant roots. The total genomic DNA of all three compartments was extracted, and shotgun metagenomic sequencing was performed by Macrogen company using Illumina HiSeqX platform (2 × 150 pair-end reads) according to the manufacturer's guidelines using the TruSeq DNA kit (Illumina). For transcriptomics, total RNA was extracted from plant leaves. Library construction and sequencing were performed by Macrogen using TruSeq Stranded mRNA Sample Preparation Kit (Illumina) and sequenced on Illumina NovaSeq6000 platform (2 × 150 paired-end settings). The analyses are currently underway, and results and conclusions will be presented at the conference.

BOUSSAID, Khadidja

Lead: Assessment of the physiological effects and heavy metal removal using a response surface approach by Phragmite australis

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In this study, the Response surface methodology (RSM) was used to describe the individual and interactive effects of two variables – initial Co concentration and initial pb concentration – at five levels.
They were combined, under controlled conditions, according to a Central Composite Design (CCD) of the duckweed Phragmite australis. Parallelly, the assessment regarding the removal of the aforementioned compounds took place. The effects of initial Co and initial Pb concentrations on the growth parameters, the photosynthetic pigment and the removal of heavy metals were part of the study.
The analysis of variance (Anova) revealed the relative significance of the process parameters in responses. Moreover, a high correlation was found between the experimental and the predicted results, reflected by R2 (coefficient of determination). This study has demonstrated that RSM can be applied to predict the physiological responses of Phragmite australis and the heavy metal removal.
The Pb and Co treatment. This study gives an insight to the possible mechanism of hypertolerance, signifying that trees can be successfully used for phytoremediation.
The method applied for this research allowed obtaining the maximum of the information counting with a limited set of laboratory experiments.

Keywords:
Phragmite australis., Biosorption ,Phytoremediation, Heavy metals, Central Composite Design, Response surface methodology

BUDIĆ-LETO, Irena

Antioxidant activity and metal micronutrient content of Croatian wines determined by electron spin resonance spectroscopy

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Wine is a source of metal micronutrients in the human diet. We explored antioxidant activities and variations in the micronutrient content in red and wine wines produced from several commercial vineyards in the Croatian coastal region of Dalmatia. Samples were selected based on different grape varieties, origins, and winemaking methods. In this study, EPR (electron spin resonance) spectroscopy was applied because the most important feature of EPR is the ability to determine the concentration of radical species, such as metal ions, or the presence and reactivity of reactive species (ROS and/or RNS). Antioxidant activity is an expression of the amount or concentration of available antioxidants, and the EPR method is based on measuring the decrease in the EPR spectrum of relatively stable radicals as a result of their interaction with antioxidants. In this study, in all wine samples, the presence of manganese (Mn) and iron (Fe) as essential metal nutrients was detected by EPR spectroscopy. In addition, the optimal conditions for EPR investigation of the antioxidant activity of wine samples were investigated to evaluate the quality of wines. A significant difference in the decrease and rate of decrease of free radicals, Tempol and DPPH, was found in red wines compared to white wines.

CALDELAS, Cristina

Main drivers of mineral concentration in wheat grains in semi-arid climate and alkaline soil.

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The aim of this study was to test the hypothesis that genetic variability is the main driver of mineral concentration in wheat grain in Mediterranean conditions. We grew 12 modern winter wheat varieties in semi-arid conditions and alkaline soils, in two consecutive years of contrasting water availability, and at three rates of N-fertilization: 64, 104, and 130 Kg N ha-1 Our results indicate that the genotype is the main driver of the mineral concentration in the grain in our conditions, especially for Ca, K, Mg, and S, but the environmental constraints can be more relevant for other minerals such as Fe and Zn. By contrast, the nitrogen fertilization rate had little effect. We attribute this to the fact that N was not a key limiting factor for nutrient acquisition in our conditions, the main issues being water scarcity or soil characteristics. We identified three potential nutritional deficiencies (P, Zn, and Fe), likely linked to the high soil pH. Furthermore, the thousand-grain weight correlated negatively with the mineral concentration in the grain, revealing the importance of grain shape. Our results support the notion that a dilution effect is a cause for the dwindling nutritional value of the grain in modern semi-dwarf wheat varieties. Among the 12 varieties tested, CH-Nara grains were highly nutritious making this variety a potential source of germplasm. To maximize the nutritional value of modern wheat varieties in Mediterranean semi-arid conditions and alkaline soils, the most effective strategy could be plant breeding, but with consideration of the local constraints (water, soil pH). The knowledge gained from this study will guide future breeding and agronomic practices in our region and contribute to guaranteeing food security in the context of climate change.

CASTILLO, Hiram

X-ray microspectroscopy at ESRF beamline ID21: applications in Plant science.

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Trace metal distribution and speciation in plants is an active research field, whether to understand metal biological roles, to improve food quality or to understand and prevent the accumulation of toxic metals in edible crops. This research domain is one of the core activities at beamline ID21 of the European Synchrotron. ID21 is a beamline dedicated to X-ray fluorescence (XRF) mapping and X-ray absorption spectroscopy (XAS) in the tender X-ray range (2-11keV), this energy range allows detecting important nutrient elements (P, S, K, Ca, Mn, Fe, Cu, Zn) as well as rare earths and pollutants (Cd, Ag, Ce, La, Gd). A brand-new X -ray nanoscope is being installed at the beamline to complement the existing microscope, and it will be soon available to users. It will offer enhanced capabilities for nano-XRF mapping, nano XAS and hyperspectral XRF mapping. This new state-of-the-art instrument will offer higher lateral resolution (down to 100 nm) with better XRF detection capacities (sub-ppm), higher acquisition speed, an improved cryogenic sample environment, preserving user-friendless thanks to a new graphical user interface. Cryo-fixed plant samples can better cope with intense X-ray beams and the elemental distributions, chemical states, and sample morphologies are close to the in-vivo state under frozen-hydrated conditions. This presentation will highlight present and future capabilities at ID21 for the plant science user community. Some examples of research done at ID21 will be used to illustrate sample preparation protocols, and data acquisition and analysis strategies.

CLEMENS, Stephan

Thioarsenates are an emerging food safety threat: accumulation, mobility, and metabolism of dimethylmonothioarsenate in rice and other plant species

Britta Planer-Friedrich, Erik Pischke, Carolin Kerl, Andrea Colina Blanco, Sebastian Haider, Fabrizio Barozzi, Stephan Clemens[1]
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Recently, both inorganic and organic thioarsenates have been found to be ubiquitously present in pore water of rice paddy soils around the world. Thioarsenates are taken up by plants and exert toxic effects (1). Transport rates and plant sensitivities vary widely. An arsenic species of particular concern for food safety is dimethylmonothioarsenate (DMMTA). Studies with mammalian and human cells consistently found a toxicity of DMMTA higher even than of arsenite. Our studies on rice and A. thaliana showed comparable toxicity profiles, as well as efficient uptake of DMMTA and high rates of translocation from roots to shoots (2, 3). Importantly, substantial amounts of DMMTA accumulate in rice grains and are detected in products derived from rice grains (4). In fact, a global survey revealed an omnipresence of DMMTA in rice grains (5). This had previously been overlooked because standard analytical procedures result in the conversion of DMMTA to dimethylarsenate, an arsenic species that is far less toxic for humans than DMMTA. Taken together, these observations call for a revision of regulatory guidelines defining the maximum tolerable levels of arsenic in food items, in particular rice (6). Moreover, there is an urgent need to elucidate the pathways of DMMTA transport and possible emergence within plant tissues. We will present our latest insights into the toxicity, transport, in planta mobility, and metabolism of DMMTA.
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COSIO, Claudia

EnCOPT1 controls Hg uptake in Elodea nuttallii

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Primary producers, i.e. aquatic plants, are instrumental for the biomagnification of the highly toxic mercury (Hg) in aquatic food webs. Nevertheless, the mechanisms of Hg accumulation remain elusive.
We identified by RNAseq a transporter involved in Hg uptake in the macrophyte Elodea nuttallii: EnCOPT1. Here, this transporter was studied by RT-qPCR in E. nuttallii and by heterologous expression in yeast, HEK293 and Arabidopsis thaliana. EnCOPT1 was proven to have a function in Cu transport by complementation of the yeast mutant ctr1Δctr3Δ. Subsequently, its activity in Hg transport was evidenced by heterologous expression in HEK293 cells. In line with these observations, Hg accumulation was reduced in loss-of-function Atcopt1 and increased in gain of function AtCOPT1 and EnCOPT1 lines in Arabidopsis thaliana. Finally competition with Cu impaired Hg uptake in HEK293 cells and A. thaliana, suggesting that Hg uptake through Cu transporters might occur in other species. Data were consistent with previous knowledge of AtCOPT1 but further suggest that this protein's function as an entry pathway for Hg into cells.
Our results point to an accidental cellular uptake of Hg via an essential metal importer, which challenges existing models that assume uptake by passive diffusion of neutral Hg.

CUI, Tuantuan

Response of rhizosphere microorganisms to heavy metal stress in hyperaccumulator plant Solanum nigrum L.

Tuantuan Cui
Sun Yat-sen University

The area around plant roots (rhizosphere) is an important hot spot for studying soil formation and nutrient cycling.
Rhizosphere microbial community structure is the result of a series of complex interactions and feedbacks among plant roots, microorganisms and soil physical and chemical environment.
Root exudates, as an intrinsic driving factor to maintain root function and vitality of rhizosphere microecosystem, are important carriers of substance exchange and information transfer in soil-plant system. Root exudates can directly affect the transfer and transformation ability and bioavailability of heavy metals at the root-soil interface.
Root exudes can change the rhizosphere microenvironment by recruiting some microorganisms, thus improving the plant's ability to adapt to the environment.
Whether hyperenrichment plants can recruit related microorganisms through root exudates to change heavy metal resistance has not been studied.

CZERNICKA, Malgorzata

Effects of zinc priming on activity of antioxidants and metabolites contents of waterlogged cucumber

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Abiotic stress is considered as major factor limiting crop yield and quality. The development of abiotic stress tolerant crops is one of the approaches to cope with this adversity, however, this approach is time consuming and requires big investments. The exogenous treatment of plants with chemical compounds can enhance abiotic stress tolerance. Chemical priming is believed to represent a promising strategy for dealing with abiotic stress in crop plants. The use of the mineral nutrients is considered one of the most promising approaches to diminish the effect of abiotic stress. The role of mineral nutrients, specifically Zn, is well understood in plant growth and development under normal conditions. However, Zn role in waterlogging tolerance is still poorly understood. The research was focused on the physiological, molecular and metabolic responses of Zn primed cucumber under waterlogging stress. The major objective of this study was to investigate the effect of foliar-applied Zn as ADOB® 2.0 Zn IDHA and ZnSO4·7H2O on waterlogging tolerance in cucumber. The H2O2 content, the activity of antioxidants such as CAT, APX, POX, GR and ascorbate (ASC) have also been studied. Using HPLC-ICP-MS QQQ and LC-MS/MS QUTRAP we identified metabolites, and biochemical pathways with a possible role in the Zn-induced priming against the waterlogging stress.

DAL CORSO, Giovanni

Relationship between metals in Noccaea caerulescens Monte Prinzera

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Environmental pollution has substantially increased the presence of heavy metals (HMs) in the biosphere, posing threats to all forms of life. Plants have evolved various degrees of tolerance to HMs, to survive and grow in contaminated areas. Worth of note is the evolution of the hypertolerance and hyperaccumulation traits. The metallicolous population Noccaea caerulescens of Monte Prinzera (MP) (Northern Appennines, Italy) is an example of Nickel hyperaccumulator.
N. caerulescens MP plants were cultivated in hydroponic culture, in nutrient solution amended with 10 µM $NiSO_4$ and submitted to different treatments: control; absence of Fe, absence of Zn, addition of 100 µM $NiSO_4$; 100 µM $ZnSO_4$ and 50 µM $CoSO_4$. After three weeks, metal accumulation was measured in both shoots and roots, and expression of genes coding for metal transporters was analyzed by Real Time PCR. Preliminary results show that increased Ni concentrations in the growth media repress Fe, Zn and Mn accumulation in leaves, while enhancing Ni and Co accumulation. In roots, 100 µM $NiSO_4$ enhanced Ni accumulation, pointing to an effect of Ni excess in reducing the root-to-shoot Fe transport rather that reducing its root uptake. The treatment with Co did not influence Ni accumulation, while reducing the accumulation of Zn, Fe and Mn in leaves.
The expression of HMA4, a plasma membrane protein involved in Zn transport, is induced in leaves and downregulated in roots by Ni excess. The expression of IREG2, involved in Ni translocation into the vacuole, is enhanced in shoots downregulated in roots of Ni-treated plants, supporting a role of IREG2 in Ni detoxification in the above-ground tissues. Fe deficiency seems to downregulate IREG2 shoots expression, while inducing its overexpression in roots. IRT1, involved in Fe absorption, was induced in roots upon both Ni excess and Fe deficiency. Further investigation will be performed on the expression of the AtFIT1 homologous, involved in Fe sensing. Also, the amount of the metal chelator nicotianamine will be analyzed.

DAUM, Diemo

Iron biofortification of spinach by means of foliar fertilization

Diemo Daum and Esther Schulz
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The study aims to develop an approach for iron biofortification of spinach, a vitamin C-rich leafy vegetable. First field trials were conducted in northern Germany, where spinach was treated with different forms of iron (iron chelates and ferrous sulfate) and fertilization rates (0.05 - 0.45 kg Fe/ha). The application was conducted by foliar sprays with a water application rate of 500 L/ha and with the addition of a wetting agent. Native iron content in spinach leaves varied between 0.7 - 1.9 mg/100 g FM depending on the site (soil pH 5.1 and 6.2, resp.) and cultivar. Within a cultivation site, the iron content of the 12 spinach varieties examined varied by a factor of 2.1. In preliminary application tests with different forms of iron fertilizer, Fe-HEDTA proved to be the least tolerated by the plants. Here, even a single treatment with 0.05 kg Fe/ha resulted in leaf necrosis. When Fe-IDHA and ferrous sulfate were applied, no leaf damage occurred with application rates up to 0.3 kg Fe/ha. Foliar sprays of Fe-EDDHA did not cause damage to leaves even when treated twice each with 0.45 kg Fe/ha. Following field trials indicate that the occurrence of leaf damage after iron foliar sprays under summer conditions is significantly affected by the time of day. Treatments in the later evening hours were best tolerated by the plants. A single foliar fertilization with 0.3 kg Fe/ha ten days before harvest approximately doubled the iron content in spinach

DEGAICHIA, Hoceme

Physiological and biochemical response of alfalfa and clover to Cobalt (CoSO4) and Zinc (ZnSO4)

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The purpose of this work is to monitor the effects of cobalt and zinc on the germination of alfalfa (Medicago sativa L.) and white clover (Trifolium repens L.) and to study some growth parameters (DNA; amylase activity), in order to determine their germinative capacity under different conditions. Germination is carried out in vitro, the seeds are set to germinate at a temperature of 25°C, in the presence of solutions at different concentrations of Cobalt CoSO4 (0.5, 0.8, 1.2 g/l) and Zinc ZnSO4 (0.7, 1, 1.5 g/l). During a duration of 14 days of experiment, physiological analyses show the effect of TM is beneficial in triggering the germination case of zinc with a germination rate increase to 90% for all concentrations used, however, the case of cobalt with reduced germination rate to 29.17% in alfalfa. TM reduces the speed of cell development and protein synthesis. In alfalfa and clover grown in the treated environment there is a decrease in the DNA content of alfalfa seeds due to metal stress of cobalt (0.8; 1.2 g/l) and zinc at (0.7; 1 g/l) Unlike clover seeds, where a high DNA content was obtained. High concentrations of Cobalt induce a low DNA content which results in sensitivity to TM. Alfalfa is therefore more sensitive to Cobalt and Zinc. The activity of α-amylase, meanwhile, is clearly accelerated by Cobalt and Zinc. Results show that alfalfa and clover seeds tolerate TM at different concentrations, however, zinc is an essential metal for germination

EROGLU, Seckin

Seeds develop dark black colour in the radicle protrusion site in the presence of ferrous in the growth media

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While working on the impact of the iron supplements during germination, we noticed Arabidopsis thaliana seeds develop an intense, reproducible black color when ferrous chloride is added to the medium. The seed germination percentage remained unaffected. Staining was localized around the radicle protrusion site and shortly preceded the radicle protrusion. Staining remained undifferentiable when several known oxidizer pathways were repressed; reactive oxygen species scavengers such as ascorbate or repressors of RBOH genes such as potassium iodide did not make any difference. Staining disappeared in the presence of gibberellic acid and expanded in the presence of abscisic acid in the medium. Similar to Arabidopsis
staining appeared also in tomato seeds, indicating the conservation of the phenomenon. We hypothesized the staining was due to chemicals in the seed coat that react with ferrous iron. A mutant that is defective in the phenylpropanoid pathway avoided being stained by ferrous chloride, confirming the hypothesis. The newly developed technique uses ferrous as a new method for staining seeds to explore seed germination dynamics.

FALKENBERG, Gerald

Biological Imaging with Synchrotron Radiation at Beamline P06 (DESY)

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The Microprobe experiment at the PETRA III beamline P06 is a versatile setup for scanning X-ray microscopy with X-ray fluorescence, X-ray absorption spectroscopy and X-ray diffraction/scattering contrasts. A KB system focusses a beam of 10^10 photons/s down to 300 nm focus size in the energy range 5 - 21 keV. By utilizing pre-focusing compound refractive lenses (preCRL) the flux can be increase by more than one order of magnitude on the expense of beamsize. Advanced detector technology, namely multi-element SDD X-ray fluorescence detectors enable on-the-fly scanning schemes with millisecond dwell times per scan pixel. The ability to collect megapixel images in less than an hour facilitates series of 2D images for full 3D fluo-tomography, spectro-microscopy, time-resolved in-situ microscopy or other multi-dimensional microscopic experiments. The microprobe setup is frequently applied for various bio-imaging applications such as element mapping of metals in soft tissues (plants, animals or human), bone, teeth or single cells. Samples can be chemically fixed, freeze dried or even fresh (unfixed). The golden standard, however, is shock-freezing and measurement in the frozen-hydrated state in order to avoid beam damage and artefacts of element re-distribution. Examples are presented for various sample types (tissues, bone, teeth, cells), scanning modes (fast 2D, tomographic 3D), and sample preparation techniques (frozen-hydrated, unfixed, chemically fixed).

FREISINGER, Eva

Metallothioneins - ubiquitous metal ion binding proteins

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Metallothioneins (MTs) form a superfamily of ubiquitous proteins that are characterized by an extraordinary large cysteine content combined with a small molecular mass (< 10 kDa). Naturally, MTs have a high affinity to thiophilic metal ions and hence function in the homeostasis of Zn(II) and Cu(I) (depending on the MT sub-form and the producing organism) as well as in the detoxification of Cd(II), Hg(II), etc. The efficiency of MTs in these processes is supported by another peculiarity: MTs are scarce in secondary structural elements such as α-helices and β-sheets. Hence the largely unfolded and highly flexible peptide chain of the metal-free (apo-) MT, also denoted as thionein, can wrap around the metal ions very efficiently and with little steric constraints. The cysteine residues are oriented towards an inner core where the metal-thiolate clusters are formed. In these clusters the metal-to-thiolate ratio is maximized by a certain fraction of thiolates that act as bridging ligands between two metal ions. In some MT forms from different phylae, mostly from plants, fungi, and bacteria, also histidine residues serve as ligands for metal ion coordination. The function of MTs is additionally linked to oxidative stress conditions brought about by the redox activity of the many Cys residues.

Although these general properties are more or less shared by all MTs, the sequence diversity of MTs from the different kingdoms of life is immense leading to variations in metal ion binding abilities and affinities as well as in their three-dimensional structures.

In the center of this contribution are the metal ion coordination properties the members of the plant MT family with an additional focus on specific peculiarities found during our research endeavours such as binding site preferences, the role of special/additional ligands, the elucidation of metalation pathways as well as the coordination of unusual metal ions such as iron in some of the sub-forms studied.

GIANNELLI, Gianluigi

Bacteria assisted phytoextraction of As by Pteris vittata grown on a naturally As rich soil

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The study of areas characterized by hydrothermal phenomena is particularly important because of the excessive exploitation of deep aquifers which lead to the mobilization of various elements including arsenic (As). The naturally As-rich soil in the Viterbo area named Bagnaccio (Lazio region, Italy) is characterized by an average As concentration of 750 mg kg−1, of which 28% is bioavailable. A high concentration of As in the hyperaccumulator fern Pteris vittata was detectable in fronds (5,000 mg kg−1), after 5.5 months of growth on Bagnaccio soil. To evaluate the possible contribution of soil microorganisms in As phytoextraction, sixteen arsenate-tolerant bacterial strains were isolated from P. vittata roots. Most of them belong to the Bacillus genus, and only two have been previously associated with As-rich soils. Six of the isolated bacteria were highly As-resistant (> 100 mM), and among them two were homologous to Bacillus simplex (PVr_2) and Bacillus halosaccharovorans (PVr_17), two others homologous to Paenarthrobacter ureafaciens (PVr_5) and Beijerinckia fluminensis (PVr_9) produced a high amount of IAA and siderophores; the last two were homologous to Acinetobacter schindleri (PVr_15 and PVr_16). Strains PVr_5 (Paenarthrobacter ureafaciens) and PVr_15 (Acinetobacter schindleri) also contained the arsenate reductase gene (ars C).
In addition, when inoculated on Arabidopsis thaliana seedlings, in the absence of As, strain PVr_9 showed an important growth-promoting effect as well as a high antifungal activity.
P. vittata plants grown on Bagnaccio soil have been inoculated with different combinations of bacterial strains: 1) the six bacterial strains together, 2) PVr_5 and PVr_15 together, 3) and individually PVr_9, PVr_5 and PVr_15. Preliminary results showed a positive effect of PVr_9, on P.vittata growth and on As phytoextraction. Further results obtained by these experiments will be presented and bacterial strains potential capacity to enhance P. vittata growth and As accumulation will be discussed.

GOMES BRITO, Fernando

Identification of a Cadmium (Cd) and Zinc (Zn) Isotope Fractionation for a Plant Metallothioneins

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Metallothioneins (MTs) are sulfur-rich proteins that bind to metal ions and are found in most organisms, including bacteria, animals, and plants. In plants, they may function separating trace metals and sequestering them in the cells of different organs such as roots and leaves. MTs are proposed to play a role in the homeostasis of Zn(II) and Cu(I) ions as well as in the detoxification of heavy metals such as Cd(II). Based on theoretical calculations for small ligands, we expect MTs to preferably bind to light Cd and Zn isotopes. Hence, isotopes could be used to study the separation of Zn, an essential micronutrient for plants and humans, and Cd, a non-essential and toxic trace metal. To date, experimental work linking MTs to the isotope fractionation of Cd and Zn in different plant parts does not exist.
We aim to determine this isotope fractionation for cicMT2 and whether the binding governs the overall isotope fractionation on a cellular level. To achieve this goal, cicMT2 was expressed in E.coli cells attached to a GST-tag for purification, which was cleaved in a second step to obtain the native protein sequence. Metal-free (apo) cicMT2 were incubated with Cd(II) or Zn(II) ions using different molar ratios and equilibration times in order to analyse the isotopic fractions of metals bound to cicMT2. After separating the unbound metal ions using size exclusion chromatography, the protein samples will be measured for isotope ratios using a multi-collector ICPMS. At the conference, we will present our data from the isotope fractionation experiment on the Cd(II) and Zn(II) uptake experiments with recombinant E. coli cells to evaluate if the isotope composition of E.coli is influenced by cicMT2. Once the isotope fractionation factor has been defined on a molecular and cellular level, we can continue to explore how isotopes can be used to study whether MTs and other sulfur-rich molecules play an important role in separating Zn(II) from Cd(II) in plants.

GUILLON, Emmanuel

Interaction between MTE and pharmaceuticals in the soil/plant system

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Human activities lead to the presence of a wide range of contaminants in the environment especially in soil and water. Among these contaminants, metallic trace elements and pharmaceuticals are often found simultaneously. It is known that they can interact together to form coordination complexes which modifies their speciation and thus affects their behavior in the environment. Indeed, their sorption properties can be modified in the presence of each other due to the formation and an influence of their simultaneous presence on their respective plant uptake can therefore be expected. There is a lack of knowledge dealing with this very often encountered case. This poster presents our methodology, through examples using a multiscale approach from macroscopic to molecular, to gain insight this issue.

HANIKENNE, Marc

A tricky journey: characterizing ZIP transporters in the hyperaccumulator Arabidopsis halleri

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Plants have the ability to colonize highly diverse environments. The zinc and cadmium hyperaccumulator Arabidopsis halleri has adapted to establish populations on soils covering an extreme range of metal availabilities. Although several ZIP transporter-encoding genes are among candidate genes that are constitutively highly expressed in roots and shoots of A. halleri, their contribution to zinc and cadmium hypertolerance and hyperaccumulation remains limited. Based on data available in the literature, we made two hypotheses, namely that: zinc deficiency-responding ZIPs are essential for zinc hyperaccumulation, in particular for root uptake and radial transfer, and (ii) ZIP6 is key for root-to-shoot zinc transfer. To challenge those hypotheses, bzip19/bzip23 (i.e. two transcription factors controlling zinc deficiency-inducible ZIP genes) and zip6 knock-down A. halleri lines, as well as overexpression lines in A. thaliana, were generated. These transgenics displayed unexpected zinc and cadmium accumulation and tolerance phenotypes: zip6 A. halleri lines showed increased cadmium tolerance, whereas bzip19/bzip23 A. halleri plants displayed increased zinc and cadmium accumulation combined with cadmium sensitivity. Based on these observations, we will discuss the functional redundancy that exists among hyperaccumulation mechanisms and how hyperaccumulation remains a suboptimal process.

HAUSER, Marie-Theres

Effects of trace metal ions on pectin methylesterase (PME)/ PME inhibitor (PMEI) expression and activity in Arabidopsis thaliana

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Cell wall acts as a bridge for signaling between the external environment and the internal cell functions. Pectin, in particular the homogalacturonans (HG) is a structural cell wall component that maintains the stability of cellulose and xyloglucans. Exposure to trace metal ions changes the composition of the cell wall, specifically pectin that plays a role in signaling external biotic/abiotic signals to the cell. HGs commonly occur in highly methylated and acetylated form in the cell wall. The degree of methylation/acetylation are controlled by 67 pectin methylesterases (PMEs) and 76 PME inhibitors (PMEIs). De-esterified carboxylic groups in HGs have the ability to exchange cations and bind apart of calcium also trace metal ions. Changes in pectin methylation levels result in the production of signaling molecules like oligogalacturonides (OGs) which further triggers physiological responses and growth adjustments. This study focuses on the effects trace metal ion exposure on PME/PMEI expression and general PME activity.

HRISTOZKOVA, Marieta

Perspectives of plant development on heavy metal contaminated soils restored by mycorrhizal fungi

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The overview of several mycorrhizal and marjoram symbiotic associations represented the effect of the studied stress over different strains and their interactions with the plants. Four mycorrhizal isolates, derived from different soils were tested: Claroideoglomus claroideum, Rhizophagus clarum, Claroideoglomus claroideum, Funneliformis mosseae. To get insight into the role of symbiosis in protecting Origanum majorana L. against the excess of metals (Cd and Pb), we investigated the growth, uptake and distribution of heavy metals in the plant parts, mycorrhizal colonization status, glomalin- related soil proteins and acid phosphatase activity (root/soil). Pb and Cd partitioning in non-mycorrhizal plants were soil>shoots>root while in symbiotic associations variants changed to soil>root>shoots. The root dry weight increased in plants inoculated by C. claroideum strain (isolated from industrially metal-contaminated sites). The highest shoot biomass correlated with the percentage of mycorrhization, relative mycorrhizal dependency, glomalin production and acid phosphatase activity was determined by C. claroideum and F. mosseae (both derived from natural metalliferous sites). The findings in this study are essential to get the most benefits of mycorrhizal association in unfavorable conditions connected with plant development and herbal products free of harmful ingredients.

CHMIELOWSKA- Bąk, Jagna

Detection of 8-OHG in root cells of soybean seedlings exposed to cadmium

Jagna Chmielowska-Bąk; Jarosław Gzyl; Joanna Deckert
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A number of soil properties, genetic potential, climate, stalk position, application of soil amendments affects the chemical composition of tobacco plants. Tobacco (Nicotiana tabacum L) is particularly sensitive to the quantities of nitrogen in soil. In the present study we evaluate the influence of increasing nitrogen rates to the chemical characteristics and trace element content of oriental tobacco varieties. Investigation was conducted on two different locations on colluvial soil type, in randomized complete block design with three replications, two nitrogen rates (20 and 30 kg/ha) with constant amount of phosphorus (60 kg/ha) and potassium (40 kg/ha) and four tobacco varieties (P-23, P-79, Basma and Elenski). Increasing content of nicotine, total nitrogen and mineral matter, and decreased content of the soluble sugars is observed in all varieties treated with higher nitrogen rate. In the group of 20 analyzed elements, higher doses of nitrogen fertilizers lead to linear increase only on the content of Ni, Fe and Co in all tobacco varieties. Zn and Cu content of the oriental tobacco leaves of all four variants decreased upon the lower nitrogen rate and decreased upon the higher dosage. The effects of nitrogen rates on the content of other elements were ambiguous, since there were uneven distributions without regularity. Compared to other varieties, it was found that Basma variety had the higher accumulation factors (BAF) for Cu, Zn, Mn and Ni.

ILYAS, Muhammad Faizan

Efficiency of foliar-applied ZnSO4 and Zn-EDTA in combination with P in maize grown under Zn deficiency

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Foliar application of zinc and phosphorus is a common practice in maize to ensure sufficient supply of these nutrients. However, mixing ZnSO4 with P results in precipitation of zinc phosphate which may compromise the application and efficiency of both the nutrients. This problem could be addressed by using chelated Zn sources. To test this possibility, three weeks old maize plants grown in Zn-deficient nutrient solution were foliar fertilized with ZnSO4 and Zn-EDTA (@ 0.03% Zn) alone and in combination with P (200 mM KH2PO4). Foliar spray solution containing KH2PO4 and ZnSO4 resulted in white precipitates, which were not observed for Zn-EDTA. A simulation model study showed that more than 20% of the total Zn got precipitated at pH below 4 when mixed solution of ZnSO4 and KH2PO4 was made. Foliar applied Zn-EDTA visibly retrieved Zn deficiency symptoms in maize plants, whereas, the effect was not prominent with ZnSO4 foliar fertilization. Compared to untreated Zn deficient plants, foliar Zn application improved SPAD values in all treatments. Application of Zn-EDTA+P to Zn-deficient maize plants increased shoot dry matter by 36%. Moreover, Zn concentration was significantly increased only by foliar fertilization of Zn-EDTA+P. Hence, it is concluded that although foliar application of all Zn sources retrieved growth of Zn-deficient maize plants but Zn-EDTA was the most appropriate Zn source for foliar application to maize in combination with P fertilizer.

IMRAN, Muhammad

Does the recipe of nutrient solution for alkalinity experiments in hydroponics need modifications?

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At high pH levels in the nutrient solution e.g., salinity or alkalinity experiments, presence of carbonates can cause severe Fe deficiency. Therefore, experiments were planned to study the ability of different Fe chelates to keep it in soluble form in the nutrient solution at different pH level due to NaHCO3.
For this purpose, nutrient solutions prepared by using different Fe-chelates with variable levels of NaHCO3 were incubated for 24 hours, 3 and 6 days at room temperature in 3 replicates. Nutrient solutions were aerated during the whole incubation period.
At the end of incubation period, nutrient solutions were analysed with ICP-OES after filtered by using MN 619 G ¼, Macherey-Nagel filter papers.
Results: After 6 days of incubation, in FeEDTA containing nutrient solution at 5 mM and 15 mM NaHCO3 concentrations, a significant increase (80.8 % and 52.2 %, respectively) in soluble Fe fraction was recorded. For FeDTPA containing nutrient solutions, at 5 mM NaHCO3 concentration, soluble Fe fraction was increased to 62.9 %, while it was slightly decreased to 42.6 % in 15 mM NaHCO3 containing nutrient solution as compared to 3 days of incubation. Iron-HBED resulted in the maximum soluble Fe fraction > 98 % at all NaHCO3 concentration levels during the whole incubation duration.

ISAURE, Marie-Pierre

Deciphering manganese loading in plant seeds: insight from a multimodal three-dimensional imaging approach combining synchrotron techniques

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Manganese (Mn) is an essential element for plants, but its availability in soils is generally low and the plants must develop some specific strategies to cope with Mn shortage and deficiency. For instance, they produce a high affinity Mn transporter from the Natural Resistance Associated Macrophage Protein (NRAMP) family at the root surface. In contrast, at high concentration, Mn is toxic and the plant needs to regulate its uptake. Mn is also crucial for seeds and germinating plants for reasons that are not very well understood. Moreover, the processes underlying Mn loading in the seeds are poorly documented. While the vacuolar Metal Tolerance Protein 8 (MTP8) transporter has been shown to play a role in Mn transport and accumulation in the embryo, other transporters of the NRAMP family seem to be involved as well.
Pioneer work on Fe and Mn in Arabidopsis thaliana mature seeds using X-ray fluorescence microtomography (XRF microtomography) proved the high suitability of the technique to probe metals in intact seeds. In this work, we carried out a multimodal three-dimensional (3D) imaging approach combining synchrotron XRF microtomography and full field X-ray microtomography to study Mn loading in the seed. This combination allows the correlation between elemental distribution and seed structure in a non-invasive way and in intact whole seeds. We particularly focused on the role of NRAMP2 transporter by investigating A. thaliana seeds and knockout mutants. Our recent results will be presented here.

JORDANOSKA SHISHKOSKA, Biljana

The effects of increasing doses of nitrogen on trace element content of some oriental tobacco varieties

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A number of soil properties, genetic potential, climate, stalk position, application of soil amendments affects the chemical composition of tobacco plants. Tobacco (Nicotiana tabacum L) is particularly sensitive to the quantities of nitrogen in soil. In the present study we evaluate the influence of increasing nitrogen rates to the chemical characteristics and trace element content of oriental tobacco varieties. Investigation was conducted on two different locations on colluvial soil type, in randomized complete block design with three replications, two nitrogen rates (20 and 30 kg/ha) with constant amount of phosphorus (60 kg/ha) and potassium (40 kg/ha) and four tobacco varieties (P-23, P-79, Basma and Elenski). Increasing content of nicotine, total nitrogen and mineral matter, and decreased content of the soluble sugars is observed in all varieties treated with higher nitrogen rate. In the group of 20 analyzed elements, higher doses of nitrogen fertilizers lead to linear increase only on the content of Ni, Fe and Co in all tobacco varieties. Zn and Cu content of the oriental tobacco leaves of all four variants decreased upon the lower nitrogen rate and decreased upon the higher dosage. The effects of nitrogen rates on the content of other elements were ambiguous, since there were uneven distributions without regularity. Compared to other varieties, it was found that Basma variety had the higher accumulation factors (BAF) for Cu, Zn, Mn and Ni.

JURKOVIC, Josip

(STSM) Metal isotopes training and networking

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The proposed Short Time Scientific Mission (STSM) has two main goals. The first goal is to acquire knowledge and training for a) Sample preparation for isotope analyses, b) concentration and isotope analyses with single collector ICP-MS and multiple collector ICP-MS, and c) evaluation and quality control of isotope data. Isotope training will be conducted with cadmium (Cd). Hands-on sample preparation training will be conducted with a) digestion of plant material using a single reaction chamber microwave system b) sample purification using resin-based anion exchange chromatography, and c) isotope spiking using 113/111Cd double spike to correct for any artifacts during sample preparation and analyses. All these steps will be conducted in certified trace metal isotope laboratories to minimize sample contamination. In addition, very clean reagents and acid cleaned working material will be used to minimize sample contamination. Analysis of Cd isotopes s divided into two steps. In a first step, purified samples will be measured using single collector ICP-MS to control for the purity of the samples by measuring Cd and elements that cause molecular (Zn, Mo) and direct interferences (Sn). In a second step, isotope compositions will be measured using MC-ICPMS in sample-bracketing mode to correct for potential plasma drifts. The second goal is networking based primarily with people from the ETH group of plant nutrition, and with researchers that are interested in bioaccumulation of heavy metals. The experience with Cd isotope training is in large part applicable to other isotopes research, which is important for the second goal. Recent study shows that silver birch bioaccumulates heavy metals in much higher concentrations than that in gold mine waste (Tl, In, Cu, Zn, Mn, Ni). The highest bioaccumulation factor was found for thallium (BCF: 25). The research of thallium bioaccumulation is quite rare and uptake mechanism is rather unknown.

KHAKUREL, Krisna

Introducing Electron Crystallography to Plant Metals community

Krisna Khakurel
ELI-Beamlines - Czech Republic (CZ)

Electron diffraction of 3D micro/nano crystals is a newly emerging field of research. The method is of special interest for solving structure of difficult-to-crystalize proteins which do not grow into high quality crystals of sizes suitable for single crystal X-ray crystallography. Further, this method can also help in precise location of the heavy metals in the proteins, and mapping charged states of the atoms. We will introduce the potential of the technique to the plant metal community so that structures of challenging membrane proteins in plant biology can be solved by this technique in the future.

KIŃSKA, Katarzyna

Insights into tellurium pollution impact over the bioavailability of selected trace metals from soil

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University of Warsaw, Poland

Unlike many trace elements, the effect of technology-critical elements (TCEs) on living organisms has not been well documented to date due to their low concentrations and lack of a meaningful industrial role in the past. This is now changing, as several of these elements emerge as essential components in developing innovative technologies – displays, semiconductors, and new energy harvesting methods. Therefore, they are becoming more and more frequently mentioned as new xenobiotics. Most compounds of Te, classified as one of the TCEs, are considered toxic. Despite this, Te can be taken up by plants, from the contaminated soil, through the root system, together with the elements necessary for their proper functioning. Even though Te is the only metalloid in group 16 of the periodic table, it shows similar properties to other elements in this family, especially Se, located just above. And so, it was observed that plants known to accumulate Se are also prone to accumulate Te. Bioaccumulation, however, is a function of the bioavailability of elements from the soil. Therefore, a necessary part of the research was to investigate interactions of Te with soil, indicating physicochemical transformations in the soil environment and determining the mobility of different Te species. To achieve this, selected Te compounds were introduced into different types of well-characterized soil with distinct physicochemical properties. Based on fractionation data after incubation with tellurium species for 1-60 days, the level of mobility of the element and its bioavailability to plants was evaluated. Subsequently, we correlated the degree of soil contamination with tellurium with the bioavailability of selected trace metals, like Cu, Zn, and Ni. ICP-MS was used to determine the total content of analytes in the solutions after digestion, as well as in the obtained extracts.

KOHANOVA, Jana

The effect of variable temperature on maize exposed to stress caused by antimony

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Antimony (Sb) causes various damages in plants, including inhibition of growth of roots and shoots, changes in physiological processes and decreases uptake of some essential elements. Antimony is released to water and soil solution mainly due to human activities, as a result of mining. The other source in the environment comes from Sb use as an agent for metal hardening, and unfortunately recently due to wars. The other serious problem for plant growth including crops and agricultural production is rising soil and water temperature increasing with air temperature often followed by droughts and salinity. This is greatly associated with climate changes. The plant root system responds to different temperature stimuli by developmental and physiological adaptation. Similarly, the presence of antimony in soil or water substrate caused growth inhibition related with some physiological and structural changes of roots. In our experiments we combined these two abiotic stresses – the presence of Sb and higher temperature. Two cultivars of maize (Zea mays) were used as experimental plants in hydroponic conditions. One of the effects of higher temperature was production of more lateral roots. Changes in the anatomy of roots and physiological processes, were observed both as the effect of variable temperature and the increased concentration of antimony in hydroponic solution. We also observed changes in the Sb uptake and translocation to the shoots. This study was supported by Slovak Grant Agency VEGA by grant VEGA 1/0472/22, by the Slovak Research and Development Agency under the contract No. APVV SK-CN-21-0034 and by the COST action CA19116 ´Trace Metal Metabolism in Plants – PLANTMETALS´.

KRÄMER, Ute

Metal hyperaccumulation and hypertolerance in Arabidopsis halleri – towards an understanding of the molecular mechanisms through cross-species comparative approaches

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The zinc and cadmium hyperaccumulator Arabidopsis halleri is an outcrossing stoloniferous perennial which is often found on soils containing toxic levels zinc, cadmium and lead. Despite these distinctive life history and extreme physiological traits, A. halleri is among the closest relatives of the genetic model plant Arabidopsis thaliana, from which it diverged between 10 and 5 mio. years ago. The close phylogenetic relationship between these two species provides an opportunity for cross-species comparative approaches aimed at addressing the genetic and physiological basis underlying metal hyperaccumulation and associated hypertolerance in A. halleri. Researchers can thus also take advantage of knowledge, resources and methodological tools uniquely available for A. thaliana. Moreover, reference genome assemblies available for several sister species of A. halleri, which are non-hyperaccumulators and exhibit merely basal metal hypertolerance like A. thaliana, can be used to strengthen comparative genomics approaches.
In our past work, we have used cross-species comparative transcriptomics in order to analyze how the regulation of gene expression is altered in A. halleri and to identify candidate genes for key roles in metal hyperaccumulation and hypertolerance. Several of these candidate genes have been functionally characterized using reverse genetics, among other approaches, by various research groups. In earlier work, we compared the genomes of A. halleri and A. thaliana using array comparative genome hybridization. The availability of a chromosome-scale genome assembly of A. halleri now greatly facilitates comparative and functional approaches in this species. We will present results from genomic comparisons as well as our work characterizing candidate gene functions. Understanding the molecular basis of metal hyperaccumulation and associated hypertolerance will be important for the future development and improvement of phytomining and phytoremediation technologies.

KUKAVICA, Biljana

Changes in activities and isoezyme profiles of antioxidant metalloenzymes in the response of plants to stress

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Superoxide dismutase (SOD; EC 1.15.1.1), catalase (CAT; EC 1.11.1.6) and Class III peroxidase (POX EC. 1.11. 1.7) are metalloenzymes with important roles in the plants antioxidant system, catalyzing the removal of reactive oxygen species (ROS). SOD catalyzes the dismutation of superoxide anion radicals to hydrogen peroxide, which decomposes to water in reaction catalyzed by CAT. POX can remove hydrogen peroxide by reacting with phenolic compounds as co-substrates. The response of plants to stressful environmental conditions (eg flooding, pathogens) are alternations in the activities and isoezyme profiles of antioxidant enzymes. Changes in antioxidant enzymes exposed to stress were examined, with regard to the stress duration, the intensity of the stress and the plants developmental stage.

KÜPPER, Hendrik

Metalloproteome factory: from crude extract to identification of metalloproteins via 3-dimensional HPLC-ICP-sfMS

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Understanding trace metal metabolism in plants is a prerequisite for improvements in food safety and quality, agricultural productivity, and environmental risk assessment (Andresen et al., 2018; Morina and Küpper, 2022). Identification of metalloproteins from crude extracts is essential to reveal response mechanisms of plants to environmental factors, as well as differences in physiology between tissues and life stages. Conventional techniques suffer from the lack of resolution and/or sensitivity. Our recent advances improved the latter in plants, algae and photosynthetic bacteria (Küpper et al., 2019). We present here a method based on three dimensions (3D) of chromatography (anion exchange chromatography followed by hydrophobic interaction chromatography and size exclusion chromatography), all based on a common buffer system that was optimised for its compatibility with the hyphenated inductively coupled sector-field mass spectrometry (ICP-sfMS, sf-ICP-MS). Using the metalloproteome of soybean roots as an example, we demonstrated that this 3D metalloproteomics could analytically purify various metalloproteins from their natural abundance in crude extracts to a quantity and purity that is sufficient for subsequent identification by ESI-MS. Selected purification pathways of metalloproteins (binding Cu, Fe, Mn and/or Zn) from the crude extract to the final identification are shown and their role in plant metabolism is discussed.

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KURT, Cemal

DETERMINATION OF MINERAL ELEMENT CONCENTRATIONS OF DIFFERENT FLAXSEED CULTIVARS (Linum usitatissimum L.)

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Flaxseed is cultivated in many parts of world for fiber, oil as well as for medicinal purposes and also as nutritional product. Flaxseed contains relatively high contents of ash (4 %), fat (30–40 %), protein (20–30 %), and dietary fiber (20 %). However, despite its high oil content, due to the potential of flax seeds to reduce the risk of certain diseases, interest in this plant has increased in recent years. Because flaxseed has an excellent nutritional profile, it has become an attractive ingredient in diets formulated for certain health benefits. In this study, it was aimed to determine the nutrient concentrations of different flaxseed cultivars. This study was conducted to assess for 9 mineral element content of 16 different flaxseed cultivars. The N, P, K, Ca, Mg, Fe, Zn, Mn and Cu concentrations of flaxseed cultivars were varied between 1.25-2.98%, 0.129-0.358%, 1.41%-1.73%, 0.32% -1.33%, 0.25%-0.51%, 7.24-65.63 mg kg-1, 45.4-76.0 mg kg-1, 32.7-81.7 mg kg-1 and 14.6-22.5 mg kg-1 respectively. When the research findings were evaluated in general, it was determined that flaxseed Kaolin cultivar had the highest and flaxseed LS Koral cultivar had the lowest concentrations of nutrients.

LE JEAN, Marie

Locked up inside the vessels: Rare earth elements are transferred and stored in the conductive tissues of the accumulating fern Dryopteris erythrosora

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Rare earth elements (REEs) are strategic metals strongly involved in low-carbon energy conversion. However, these emerging contaminants are increasingly disseminated into ecosystems, raising concern regarding their toxicity. REE-accumulating plants are crucial subjects to better understand REE transfer to the trophic chain but are also promising phytoremediation tools. In this analysis, we deciphered REE accumulation sites in the REE-accumulating fern Dryopteris erythrosora by synchrotron X-ray µfluorescence (µXRF). This technique allows a high-resolution and in situ analysis of fresh samples or frozen-hydrated cross sections of different organs of the plant. In the sporophyte, REEs were translocated from the roots to the fronds by the xylem sap and were stored within the xylem conductive system. The comparison of REE distribution and accumulation levels in the healthy and necrotic parts of the frond shed light on the differential mobility between light and heavy REEs. Furthermore, the comparison emphasized that necrotized areas were not the main REE-accumulating sites. Finally, the absence of cell-to-cell mobility of REEs in the gametophyte suggested the absence of REE-compatible transporters in photosynthetic tissues. These results provide valuable knowledge on the physiology of REE-accumulating ferns to understand the REE cycle in biological systems and the expansion of phytotechnologies for REE-enriched or -contaminated soils.

LETORT, Fabien

Cadmium pathways in agronomic cultivars of Theobroma cacao and effect of micronutrients

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The new European regulation on the cadmium (Cd) limit of 0.6 mg Cd.kg-1, in cacao products1, affects the cacao market worldwide, especially in Latin America. Indeed, 30 to 50% of cacao bean production in this region exceeds this limit. Research on strategies to reduce Cd accumulation in cacao beans is currently limited by a lack of understanding of Cd transfer pathways in cacao trees2. Recent studies on native cacao trees3,4 have taught us more about the transfer of Cd from soil to beans. In this study, we focused on agronomic cultivars representative of production in Ecuador (CCN-51 and Nacional). We studied the effect of micronutrient deficiency on the absorption of Cd and calcium oxalate crystals (CaOx) on their possible detoxification role. We characterized total and available metal concentrations in soils, total metal, and oxalate concentrations in plant organs by inductively coupled mass spectrometer (ICP-MS) and by high pressure ion chromatography (HPIC), and the observation and analysis of CaOx under a scanning electron microscope (SEM). The plant Cd concentrations were 17 to 24 times higher than the topsoil total Cd and increased as: nib<root< mature leaf<young leaf<branch. Cd accumulation appears to be related to soil deficiencies in micronutrients such as iron (Fe), nickel (Ni), manganese (Mn), and zinc (Zn). The study of CaOx made it possible to learn about their distribution in the roots and branches and for the first time on their dosage in all the organs of the cacao trees studied. However, an assumed detoxification role has not yet been elucidated.

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LIAO, Feixue

The Zinc Deficiency Response in Zinc Hyperaccumulator Species

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Zinc (Zn) is an essential element for all living organisms. However, Zn-deficient soils are widespread globally and Zn-deficient human diets affect about one-third of the world population. The Zn deficiency response in Arabidopsis thaliana is regulated by bZIP19 and bZIP23, two F group basic region leucine-zipper (F-bZIP) transcription factors which are also sensors of intracellular Zn concentration through their Zn-Sensor Motif (ZSM). Mutations in the ZSM of the A. thaliana bZIP19 lead to an increment in plant and seed Zn contents, suggesting that F-bZIPs might be an attractive target for Zn biofortification. Considering that Zn hyperaccumulator species naturally accumulate high concentrations of Zn in their leaves, it will be interesting to understand the F-bZIP-regulated Zn deficiency response in hyperaccumulator species and whether it can contribute to Zn biofortification in non-hyperaccumulator crops. Here, we identified and functionally characterized the F-bZIP homologs from two model hyperaccumulator species, Arabidopsis halleri and Noccaea caerulescens. Results indicate that the A. halleri AhbZIP23 and the N. caerulescens NcbZIP19/23 are the functional homologs of A. thaliana bZIP19/23. A phylogenetic analysis of Brassicaceae F-bZIPs reinforces their evolutionary conservation. The possibility that the F-bZIP-regulated Zn homeostasis might be involved in the Zn hyperaccumulation trait in A. halleri is discussed.

LICHTSCHEIDL, Irene

Zinc and the interplay between Arabidopsis arenosa and endophytic yeasts

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Arabidopsis arenosa is a plant species able to grow in soils that are heavily contaminated with heavy metals (HMs). Zinc enters the roots but it is not hyper-transported to the above-soil organs. When grown in vitro on zinc rich substratum, endophytic yeasts Sporobolomyces ruberrimus and Rhodotorula mucilaginosa start to grow on the surface of the roots, forming a dense sheath. Root hair growth is severely affected, and yeasts proliferate around exploded tips of root hairs.

We investigated the tolerance of these wild-type yeasts to zinc and we localized zinc by zinc-sensitive fluorescent dyes Fluozin-3 and Newport Green DCF as well as by Electron Spectroscopic Imaging (ESI) in the Transmission Electron Microscope (TEM).

Photometric turbidity showed that yeast cells tolerated concentrations up to 10-4 M zinc in the medium without reduction of number. The size of individual cells increased slightly in 10-5 and 10-4 M zinc, and cell walls became thick. Microscopic analysis showed that the cytoplasm filled with small vesicles and finally coagulated. The fluorescent indicator dyes localized zinc in young living cells to cell walls, cytoplasm and developing buds, but not to the large vacuole. With high concentrations, zinc accumulated in the small additionally formed vesicles and remained there also after cell death as nano-crystals. ESI in TEM proved these localizations and indicated in addition the presence of nitrogen and sulphur in these nano-crystals, suggesting the contribution of proteins during formation. The same results were observed for yeast cells that had developed inside roots and root cells.

There is increasing evidence for various mechanisms of endophytic yeasts to improve plant growth; our results suggest that endophytic yeasts have the potential to act as biological filters that reduce the toxic load of zinc in and around plant roots. Further investigation is needed to understand cost/benefit relations of the plant/endophyte interplay.

LUX, Alexander

Little known function of root cap in protection of plants against toxic metal concentration

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Root cap is mainly known as a part of root controlling root gravitropism and facilitating root penetration through the soil. It is known also as one of the plant checkpoints. Root cap protects the most sensitive root part – root apical meristem against mechanical injury, and various abiotic and biotic stresses. Root cap can also modify the uptake of some trace elements to the root and its translocation to the stem, leaves, and generative organs, as we have shown in one of our previous studies. In this contribution we focus on boarder cells. Boarder cells are released from the surface of the root cap in land plants and contribute to the interaction of the root surface with the soil participating to the continuum known as rhizosphere. We have studied their role in modifying uptake and translocation processes of nickel. Nickel in low concentrations is an essential element, without which plants cannot complete their life cycle. Due to its useful properties Ni is widely used also in modern technologies. Nickel deposits have been exploited in many areas of the world leaving large areas of Ni-rich substrates with negative impact on plant growth, crop yield and human health. In our study we have shown that boarder cells are important in regulation of Ni transport to the roots and leaves of maize (Zea mays) cultivated in aeroponics. Changes in structure and metabolism of plants accompanied this effect. This study was supported by the Operation Program of Integrated Infrastructure for the project, UpScale of Comenius University Capacities and Competence in Research, Development and Innovation, ITMS2014+: 313021BUZ3, co-financed by the European Regional Development Fund, by Slovak Grant Agency VEGA by grant VEGA 1/0472/22 and by the COST action CA19116 ´Trace Metal Metabolism in Plants – PLANTMETALS´.

LY, Serigne

Physiological response in relation to nickel hyperaccumulation in Bornmuellera emarginata cultivated in hydroponics over a nickel gradient

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The nickel hyperaccumulator plant Bornmuellera emarginata (Brassicaceae) is a wild species highly adapted to thrive on nickel-rich ultramafic soils in the Balkans. The population from Euboea Island (Greece) was subjected to an ecophysiological investigation to determine responses to exposure to different nickel concentrations (0, 1, 10 and 100 µM Ni) in hydroponic solution. Nickel hyperaccumulation (1000 mg kg-1) was confirmed in this population and occurred at a dose rate of 10 µM in hydroponic solution. Foliar nickel accumulation was strongly correlated (r = 0.99) to the nickel concentration in the solution. The results showed that the Translocation Factors and Bioconcentration Factors were much greater than 1, even in the lowest exposure level of 1 µM Ni. This confirms the enhanced ability of B. emarginata to accumulate nickel over a wide concentration range. The results show that nickel exposure up 100 µM Ni induces only mild physiological stress symptoms. Bornmuellera emarginata is shown to reduce the relative water content of the leaves in response to nickel, as this parameter was significantly different from the control (0 µM). This mechanism facilitates this species to maintain the same biomass (of the leaves and roots) in all of the nickel treatment levels, which did not differ significantly for this parameter. The potential for this species to accumulate Zn could be explored (as well as interactions between Ni and Zn).

MAKSIMOVA, Viktorija

Evaluation of heavy metals accumulation potential in dry flower of different Cannabis varieties

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Presence of heavy metals in the ecosystems in soil and plants, both can be harmful for human health. Determination of heavy metals in the varieties of Cannabis for medicinal use is of importance since their extracts can be used in preventive or medicinal aims. Cannabis sativa varieties have many purposes, and in the recent studies are even found as one of the possible biological methods for decontamination of the soil from trace metals. In this study we evaluate the Cannabis varieties accumulation potential for some trace elements that are strictly controlled.

Six different varieties of Cannabis sativa (Jack Kush, Charlotte's Angels, Mazar, Glueberry OG, Gollosa and Big Food Glue) have been germinated in the same indoor conditions and their clones have been planted in a green house. Pb, Hg, As, and Cd content in dry flowers were analyzed by internal validated GS-MS method. Highest concentrations of Pb (0.035 mg/kg and 0.045 mg/kg, accordingly) were detected in Mazar and Glueberry OG varieties; Hg (0.035 and 0.02 mg/kg) and As (0.119 and 0.260 mg/kg) in Jack Kush and Glueberry OG; and Cd (0.084 and 0.055 mg/kg) in Jack Kush and Gollosa, respectively. The results lead to a conclusion that Jack Kush and Glueberry OG varieties have the highest possibility for accumulation of heavy metals, especially arsenic, with highest accumulation rate.

MAKSIMOVIĆ, Ivana

Chemical composition of winter and spring oilseed rape (Brassica napus L.) in the presence of nickel

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The oilseed rape is the most important plant for oil production in Central and Western Europe, including biofuels industry. It provides the earliest spring and the latest fall green fodder and the first bee pasture. Since it has a short vegetation period, it enables timely and high-quality preparation of the soil for the next crop and allows two harvests per year.
The experiment was set up in a greenhouse under semi-controlled conditions. Before sowing, the seeds of one spring and one winter oilseed rape cultivars were immersed for 24 hours in a 10 µM NiSO4. Plants were grown on Hoagland solution containing 10 µM NiSO4 for a month, after which their mineral composition was analyzed.
Nickel content was many times higher in plants exposed to Ni. The concentration of P, Mg and Zn was 34-60% higher than in the control plants, while the content of K and Ca varied depending on the organ and variety. The content of Mn, both in the leaves and stems was lower compared to the control by up to 53%. The concentration of B in leaves was 11-22% lower than in the control, whereas in stems of winter variety it was 2 times higher. Concentrations of Cu and Fe were lower in leaves (up to 16%), and higher in the stems (up to twice) compared to the control.
These results suggest that even if low concentrations of Ni may not affect strongly the growth of plants, they may significantly change their mineral composition, which is very important considering especially its use for fodder.

MENCH, Michel

Application of phytomanagement strategies in contaminated areas of the SUDOE space

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Phytomanagement is a set of phytotechnologies combining (1) phytoremediation options based on the use of plants (trees, shrubs, and herbaceous) and associated microorganisms to control the pollutant linkages due to contaminant excess in soils at degraded sites, (2) the production of valuable biomass that can be locally processed to supply e.g., wood, resin, essential oils, bioenergy, ecomaterials, biosourced chemicals, ecocatalysts, etc., and (3) the remediation / supply of ecological functions to enhance ecosystem services (e.g., C sequestration, erosion control, creation of habitats, and biodiversity conservation).
The Phy2SUDOE project aims to value sites contaminated by-metal(loid)s and organic compounds in the Southwest European region (SUDOE) through the phytomanagement use. This project aims also to preserve the endemic biodiversity typical of some contaminated sites (e.g., metallophytes, plant growth-promoting bacteria, mesofauna, etc.) due to their intrinsic and utilitarian value (e.g., biotechnological applications) and role in the ecosystem life cycle. The PhytoSUDOE network (https://www.phytosudoe.eu/en/) has been extended to 15 contaminated sites distributed over Southwest France, Portugal and Spain, with the addition of new case studies, phytomanagement strategies, partners and stakeholders. The human capital of the network has been expanded with various partners (site managers, universities, R&D centers, companies, and administrations) to stimulate the creation of solutions and management strategies and the result transfer. Each site has its own action plan and management: i.e. conceptual model, feasibility of options, remediation strategies, protocols, monitoring, etc., while following harmonized practice guidelines. The 15 sites total an area of 350,000 m² where the various phytomanagement strategies are applied. The diversity of the site properties, as well as the surface area treated, makes it possible to improve the ecological quality of these SUDOE areas, but also to develop protocols, tools and management models that can be implemented in many contaminated sites that exist today in the SUDOE zone and

in the world. This presentation will highlight examples of phytomanaged sites in Nouvelle-Aquitaine, Occitanie, Basque Country, Galicia, and Northern Portugal.

MESTROT, Adrien,

Increasing temperature and flooding enhance arsenic biomethylation and biovolatilisation in Swiss soils.

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Reductive dissolution is one of the main reasons for arsenic (As) mobilisation in flooded soils while biomethylation and biovolatilisation are two microbial mechanisms that greatly influence the mobility and toxicity of As. Climate change will lead to more extreme weather events such as flooding and higher temperatures, potentially leading to an increase in As release and biotransformation. Here, we investigated the effects of flooding and temperature on As release, biomethylation and biovolatilisation from two As-rich soils with different pH and As origin. Namely, one acidic, mine impacted Umbric Leptosol from Salanfe (VS) and one pH-neutral, geogenically impacted Calcaric Cambisol from Liesberg (BL). Flooded soils incubated at 23°C for two weeks showed an increase of 87% (Liesberg site) and 48% (Salanfe site) in total As concentration of soil solution compared to those incubated at 18°C. Methylated As and thio-As species were found in acidic soil and soil solution. Elevated temperature enhanced thiolation and methylation although inorganic As remained dominant. We also show that volatile As fluxes increased more than 4-fold between treatments, from 18 ± 5 ng/kg/d at 18°C to 75 ± 6 ng/kg/d at 23°C from Salanfe soil. Our results suggest that high As soils with acidic pH can become an important source of As to the surrounding environment according to realistic climatic scenarios and that biovolatilisation is very sensitive to increases in temperature. However, further investigations in more sites are needed to generalise this statement and these should also include microbial analyses as well as solid-state speciation to better understand the role of S and Fe biogeochemistry in this context.

MILIĆEVIĆ, TIJANA

Environmental implication, human health risk and bioaccessibility assessments based on potentially toxic elements in vineyard soil and grapes

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In agricultural environments, frequent application of agrochemicals and activities from surrounding pollution sources directly leads to increase concentrations of pollutants in soil and grapevine. We performed several experiments in different vineyards (conventional and organic) in Serbia. The concentrations of potentially toxic elements (PTEs) were investigated in soil, soil bioavailable fractions, grapevine, and gastrointestinal extracts of soil and grape samples. These experiments were conducted to assess element mobility and bioavailability in the soil−grapevine system and examine environmental implications, the worst-case scenario of workers' and consumers' health risk and the bioaccessibility of PTE in the human gastrointestinal tract (GIT). Performing in vitro unified bioaccessibility method (UBM) for element concentrations available from soil and grape in human GIT was assessed. Environmental risk assessments pointed out the most polluted locations between investigated vineyards. Human health risk assessment (worst-case scenario) showed some risk for workers but there were also risk and moderate risk for consumers of some grape varieties. It was identified that Cr and Ni concentrations in soil and grape mostly contributed to environmental implications and increasing the human health risk. The highest contribution to risk for workers had pseudo-total concentrations of Ni, then Cr, Co and Mn from the soil, and for consumers Ni, Cr, Mo and B concentrations from grapes. However, these elements from soil and grapes were negligibly bioaccessible in human GIT assessed by the UBM bioaccessibility test, but anyhow the worst-case scenario risk assessment can be used as a precautionary measure in vineyard management. Also, Cr and Ni were highly bonded in residual soil fraction and they were not mobile, while Co and Mn were bonded in reducible soil fraction. Easily available were Mo and B from the soil, but they were negligibly bioaccessible to human. Finally, PTEs investigation in samples from vineyards accompanied by environmental and human health risks with bioaccessibility assessments may improve vineyard production and management.

Misljenovic, Tomica

New discoveries of metal hyperaccumulation in the Balkans

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Herbaria represent an important source of information about the Plant Kingdom. Novel methods provide new avenues for nondestructive analysis of herbarium material, such as the analysis of plant ionomes using portable XRF instruments. The analysis of ionomes can lead to the discovery of new metal hyperaccumulators, which is a very rare phenomenon, found only in about 2% of vascular plants. The peculiarities of the Balkan Peninsula make it one of the centres of European plant diversity, and the particular geology of the area with a significant occurrence of nickel-bearing ultramafites contributes to the presence of a large number of Ni hyperaccumulators. The largest collections of plant material from the Balkans are deposited in the Herbarium of the University of Belgrade (BEOU) and in the collection of the Natural History Museum in Belgrade (BEO). The collections of these two herbaria were systematically scanned in order to analyse the ionome of plants and to determine hyperaccumulation in certain taxa, paying special attention to the representatives of the Brassicaceae, Violaceae, Euphorbiaceae and Violaceae.
Of the more than 3000 herbarium specimens scanned, hyperaccumulation of Ni was confirmed in a number of representatives of the Brassicaceae family (Noccaea, Bornmuellera, Odontarrhena), as well as in several specimens of Viola dukadjinica. Hyperaccumulation of zinc was detected in the species of Noccaea and Cardaminae, while simultaneous hyperaccumulation of Ni and Zn has been recorded for the first time in N. praecox. An unusual accumulation of Mn was noted in specimens of Euphorbia glabriflora, and hyperaccumulation of Tl was detected in representatives of the genus Viola, endemic to the area of Allchar (North Macedonia). Further systematic scanning of herbarium collections in the Balkan Peninsula could contribute to the discovery of new hyperaccumulators of metal(oids) with potential for application in biotechnology, as well as to a better understanding of the complex phylogenetic relationships of certain taxa in the Balkans.

Mišúthová, Adriana

How arsenic affects activities of important enzymes involved in the phenylpropaniod pathway in maize grown with silicon

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One of the biggest problems in the world today is environmental pollution. The impact of abiotic stress on plants is one of the biggest challenges of current research in plant physiology. Both arsenic (As) and silicon (Si) are metalloids - although the first one is toxic in general, the latter one is considered as beneficial for plants suffering from various kinds of stress. The aim of our work was mainly to compare two hybrids of Zea mays (L.) Tweetor (higher drought tolerance) and Luciana (sensitive to drought) which were exposed to toxic As alone (75 uM or 150 uM) or in combination with Si. We focused mainly on the shoots (specifically the first and the second leaf which were separated to blades and sheaths). We mainly monitored the activity of enzymes involved in the phenylpropanoid pathway – polyphenol oxidase (PPO) and peroxidase (G-POX), and selected related metabolites, like anthocyanins and total phenolics. The results showed that the activity of PPO and also G-POX was significantly higher in the hybrid Tweetor in As treatments, and Si affected the activity of these enzymes. Both anthocyanins and phenolics occurred to a higher extent mainly in the Tweetor hybrid, but anthocyanins were most abundant in the leaf sheaths and phenolics in the blades. The concentration of anthocyanins and phenolics increased with increase of As, and Si reduced their accumulation. From our results, we assume that increasing the activities of important enzymes in Tweetor hybrid may be one of its defense mechanisms against toxic arsenic, which is probably also related to its higher tolerance to drought. This work was supported by the Grant Slovak Research and Development Agency under the contract No. APVV-17-0164, APVV SK-CN-21-0034, Grant Agency VEGA, No. VEGA 1/0472/22 and by Grant of Comenius University in Bratislava for young researchers UK/232/2021 and UK/190/2022.

MORINA, Filis

Revealing the role of manganese and zinc in mite galls induced on the leaves of Tilia cordata

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Galls are remarkable examples of biochemical, physiological and morphological changes of plant organs induced by various organisms. Gall-inducing mites are economically important pests of grasses, agricultural and ornamental crops. They can considerably affect the plant physiology and production. Gall-mite feeding induces local cell hypertrophy and tissue hyperplasia, as well as changes in the chemical composition by yet unknown mechanisms. Several species from the family Eriophyidae induce nail galls and patch erinea specifically on the leaves of linden.
In the current study, high resolution µXRF tomography of intact frozen-hydrated galls showed that Mn in nail galls accumulated in secretory cells, idioblasts, while in erinea it was mostly localized in the cell walls of trichomes at the abaxial leaf side. Besides Mn, we revealed redistribution of Fe from palisade cells to idioblasts, and Zn and Cu to the nutritive tissue indicating specific functions of these elements. MicroXANES tomography showed that Mn complexation differed the most in the nutritive tissue compared to Mn-loaded idioblasts, parenchyma or epidermis. EPR analyses showed increased accumulation of Mn2+ bound to proteins and LMW ligands in the nail galls, as well as higher affinity of isolated erinea cell walls for Mn binding. Transcriptomic analyses showed upregulation of germin-like proteins (Mn cofactor) in both type of galls, as well as upregulation of several metal transporters, while HPLC-ICP/MS analyses showed accumulation of Mn and Zn in the membrane-bound proteins in the nail galls. The aims of our ongoing work are to reveal metal-binding proteins and low molecular weight compounds in different tissues of galls and to determine their function, whether (and which) metals accumulate to accommodate the mite development and reproduction, or they are part of plant defence response.

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MUKARRAM, Mohammad

Mycoremediation of Trace Elements

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Environmental pollution caused by trace elements poses significant challenges to ecosystems and human health. Traditional methods for remediating trace element-contaminated sites often involve costly and intrusive techniques. In recent years, mycoremediation has emerged as a promising and sustainable alternative for the remediation of trace element contamination. Mycoremediation utilizes the unique capabilities of fungi to sequester, transform, and detoxify trace elements, thereby reducing their bioavailability and environmental impact. Fungi, such as certain species of mushrooms and molds, possess specialized enzymes and metabolic pathways that enable them to degrade or convert toxic trace elements into less harmful forms. The present study aimed to utilize mushrooms for trace element remediation. We attempts to explore the mechanisms by which fungi interact with trace elements, including their abilities to accumulate, immobilize, and transform these contaminants.

The effectiveness of mycoremediation suggests its application in various contaminated environments, such as mine tailings, industrial waste sites, and agricultural lands. We present data on the efficiency and long-term sustainability of mycoremediation approaches, considering factors such as fungal species selection, environmental conditions, and trace element concentration.

NADEEM, Muhammad Azhar

Genome-Wide Association Study Unveils Genomic Regions Associated with Seed Magnesium Contents in Turkish Common Bean Germplasm

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Magnesium (Mg) is the fourth most abundant element in the human body and plays the role of cofactor for more than 300 enzymatic reactions. In plants, Mg is involved in various key physiological and biochemical processes like growth, development, photophosphorylation, chlorophyll formation, protein synthesis, and resistance to biotic and abiotic stresses. A total of 183 common bean accessions collected from 19 provinces of Turkey were used as plant material. Overall mean Mg contents for both environments varied from 0.33 for Nigde-Dermasyon to 1.52 mg kg−1 for Nigde-Derinkuyu landraces, while gross mean Mg contents were 0.92 mg kg−1. Marker-trait association was performed using a mixed linear model (Q + K) with a total of 7,900 DArTseq markers. A total of six markers present on various chromosomes (two at Pv01, and one marker at each chromosome i.e., Pv03, Pv07, Pv08, Pv11) showed statistically significant association for seed Mg contents. Among these identified markers, the DArT-3367607 marker present on chromosome Pv03 contributed to maximum phenotypic variation (7.5%). Additionally, this marker was found within a narrow region of previously reported markers. We are confident that the results of this study will contribute significantly to start common bean breeding activities using marker assisted selection regarding improved Mg contents.

OUSTRIERE, Nadège

Phytomanagement strategies for a metal-contaminated agricultural soil to provide biomass for clean biofuel production – Experience from a field trial

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The use of humic and fulvic acids in combination with mycorrhizae to increase biomass production of lignocellulosic crops grown on metal-contaminated soils is still not sufficiently documented at field scale.
This field study assessed the yield and metal uptake of Miscanthus x giganteus (miscanthus), Cannabis sativa L. (hemp), and Sorghum bicolor (sorghum) grown on a metal-contaminated agricultural soil (14 mg Cd, 731 mg Pb and 1000 mg Zn kg−1) amended with either humic/fulvic acids (L) or humic/fulvic acids and mycorrhizae (MxL), or without amendment (C).
After a 5-month growth period, the number of plants, and fresh and dry biomass of aboveground plant parts were measured. Metal concentrations in the aboveground plant parts and total and Ca(NO3)2-extractable soil metals were determined.
Results showed that the number of plants and total dry weight yields of sorghum and hemp did not significantly differ between the treatments, although an increasing trend was observed for the MxL treatment in sorghum and a decreasing trend for hemp. The shoot concentrations of Cd, Pb, and Zn for hemp and sorghum did not significantly differ between the treatments. However, an increasing trend occurred for the MxL treatment regarding stem Cd, Zn and Pb concentrations of hemp and sorghum while a decreasing trend was found for the leaves of hemp and the inflorescence of both hemp and sorghum as compared to the control. The Miscanthus was not harvested due to the drought period experienced after the rhizome transplantation, which prevented them to well sprout.
Results from a second year will clarify if the MxL treatment is a relevant one for producing energy-crops on such metal-contaminated soil.

PEITER, Edgar

Metal Tolerance Proteins - versatile key players of metal homeostasis in model plants and crops

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Proteins of the Cation Diffusion Facilitator (CDF) family, called Metal Tolerance Proteins (MTPs) in plants, mediate the efflux of metals from the cytosol for detoxification, storage, and distribution. Current understanding of the physiological roles of those transporters is largely restricted to model plants, and the roles of numerous members of this family are still obscure. Our work identified physiological circumstances that require the activity of MTPs in the model species Arabidopsis thaliana. For instance, the vacuolar AtMTP8 is strongly upregulated in roots upon Fe deficiency and serves to sequestrate Mn, which otherwise would inhibit the Fe acquisition machinery. AtMTP8 furthermore serves to store Mn in the embryo for efficient germination, and it confers the interim storage of mobilized Fe during germination. Another Mn- and Fe-translocating AtMTP member operates in xylem loading and the redistribution of Mn and Fe in roots and leaves. Our recent studies on two crops, Lupinus albus (white lupin) and Beta vulgaris (sugar beet), revealed that the toolbox of MTPs is employed with a high versatility that cannot be directly inferred from the Arabidopsis model. In L. albus, large amounts of Mn are accumulated in leaves due to rhizosphere alterations upon P deficiency. Unlike in Arabidopsis, this high Mn load transcriptionally induces LaMTP8.1 in leaves, conferring the sequestration of Mn in mesophyll vacuoles. A characterization of all MTPs in B. vulgaris, including subcellular localization, substrate selectivities, and transcriptional regulation upon exposure to metal deficiencies and toxicities, revealed unexpected deviations from their Arabidopsis counterparts. For instance, in contrast to Arabidopsis, Mn- and Zn-sequestrating BvMTPs were not induced in Fe-deficient roots, pointing to differences in the Fe acquisition machinery, while high Zn load caused a massive upregulation of Zn-BvMTPs. The results suggest that the employment of the CDF toolbox is highly diverse amongst dicots.

POBLACIONES, Maria J.

Zinc biofortification in broccoli and broccolini: Effects on plant growth and mineral composition

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Agronomic zinc (Zn) biofortification of crops could help to alleviate dietary Zn deficiency, which is likely to affect more than one billion people worldwide. To evaluate the efficiency of agronomic Zn biofortification in the Brassica genus, broc¬coli and broccolini – a new hybrid crop variety derived from a cross between kalian cabbage and broccoli were tested with four different treatments: i) no Zn application (Control); ii) soil application of 5 mg kg-1 ZnSO4•7 H2O (Soil); iii) two sprays (15 mL pot-1 each) of 0.25% (w/v) ZnSO4•7 H2O in broccoli and an unique spray (15 mL pot-1 each) of 0.25% (w/v) ZnSO4•7 H2O application in broccolini (Foliar); iv) and soil + foliar combination (Soil+Foliar). In broccoli, the florets were harvested at maturity but in broccolini, florets were harvested in four sequential harvests, being a decrease in both growth and leaf composition of Zn, Ca, Fe and Mg. Soil Zn ap¬plication increased Zn-DTPA concentration by more than 3.5-times in both crops, but did not affect plant Zn concentration or plant growth except in broccolini, where the floret production was lightly increased. In broccoli, foliar application increased Zn concentration in stem+leaves and florets of 12- and 2.5-fold in Foliar and Soil+Foliar treatments, respectively, with decreased PA:Zn molar ratios, and in broccolini, foliar Zn application increased stem + leaves and floret Zn concentration by 78 and 23 mg Zn kg-1, respectively, with a better bioavailability. Boiling reduced Zn concentration by 40% and 19% in broccoli and broccolini, respectively, along with a decrease of other mineral nutrients but increased bioavailability by decreasing the phytic acid concentration. Therefore, both entire plants could constitute a good nutritional source for animals and humans not only for Zn but also for other nutrients as Ca, K, Mg, P, S, Fe, Mn and Se.

PODAR, Dorina

EXPLOITING PLANT RHIZOSPHERE INTERACTIONS FOR RESTORATION OF MERCURY CONTAMINATED SOILS

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Mercury (Hg) pollution is a global threat to human and environmental health because of its toxicity, mobility and long-term persistence. Although Hg production has dropped, former mines and chloralkali plants represent constant sources of Hg pollution. Highly costly engineer-based technologies are used for Hg heavily contaminated areas, but they are not suitable for agricultural or extensive contaminated soils. Phytomanagement approaches using plants and rhizosphere-associated microorganism can be employed to develop safer and long-term sustainable strategies for restoring soil health and resilience. Within the current research we focused on i) assessing the metal resistance strategies of native plants colonizing a polymetallic Hg contaminated site and on ii) identifying rhizosphere-associate microorganisms (bacteria and fungi) that can assist plant growth under Hg contamination. Ruderal plant species, inhabiting a highly Hg contaminated sites, were screened for their Hg and other metals concentrations and classified according to their phytoremediation traits. The bacteria and fungi biodiversity of the plants' rhizosphere was evaluated for it could provide insights into the feasibility of a microorganism-assisted phytomanagement strategies. Cultivable rhizosphere microorganisms, bacteria and fungi, were assessed for their phenotypic resistance to Hg and to other metal contaminates. Hg-resistant bacteria were further screened for their ability to produce plant growth promoting compounds such as auxin, siderophores and ammonia. A selected plant species was investigated on its own or in association with a bacterium, a fungus or a combination of the two microorganisms, for its capacity to grow and possibly decontaminated a Hg polluted soil. The Hg-resistance mechanism of the fungi is further investigated.

PONGRAC, Paula

Iron and zinc partitioning, grain allocation and local chemical environment in proso millet (Panicum miliaceum L.)

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Millets are a botanically diverse group of small grain cereals whose agricultural importance is regionally-specific. Proso millet (also referred to as common and broomcorn millet) has historically been consumed in Central and Eastern Europe, where its remarkable drought tolerance accompanied by short growth season, minimal agronomic input requirement and nutritious grain are particularly valued. No information is available for iron (Fe) and zinc (Zn) concentration and their bioavailability in proso millet grain. In this study, a cultivar and six landraces proso millet from across Slovenia were grown in a pot experiment until grain maturity. Concentration of Fe and Zn was determined in root, shoot and grain using energy-dispersive-X-ray fluorescence. Tissue-specific allocation of Fe and Zn in grain was determined using micro-particle-induced X-ray emission. In grain embryo Fe and Zn local chemical environment was assessed using micro-X-ray absorption near edge spectroscopy. The bulk concentration (mg kg-1 dry weight) ranges in root, shoot and grain were, respectively: 102-617, 52.3-221 and 65.1-139 for Fe and 2.9-75.9, 13.6-65 and 24.3-60.1 for Zn. On average, there was smaller variability in grain Fe than in grain Zn. There was no significant difference in the bulk grain Fe concentration between the proso millets studied. By contrast, grain Zn concentration of landrace Odranci (with average 32.4 ± 1.18) was significantly smaller than of cultivar Soncek (with average 40.7 ± 1.83), therefore they were selected for further in-depth analyses. Results from tissue-specific analysis confirmed the bulk differences in grain Zn concentration and a specific grain tissue, the scutellum, contributed most to those differences. In grain embryo, 71.3% of Fe was bound to phytate and the remaining 29.7% to non-phytate ligands (i.e. citrate), while Zn was bound to phytate (45.8%), cysteine (35.3%), histidine (15.7%) and citrate (3.22%). Differences in Fe and Zn localisation and ligands in the grain of the two studied proso millets will be discussed and compared to other (pseudo)cereals in terms of Fe and Zn bioavailability.

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PUCAR, Ivana

Relationship between the activity of Class III peroxidases, the concentration of phenolic compounds and the content of heavy metals in Halacsya sendtneri (Boiss.) Dörfl. (Boraginaceae)

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Halacsya sendtneri (Boiss.) Dörfl. (Boraginaceae) is a type species of the genus Halacsya Dörfler, exclusively found in serpentine soils. Our aim was to determine the relationship between the class III peroxidases (POX), phenolics (PhOH) and heavy metals (HM: Cu, Fe, Ni, Cr) in the leaf and rhizome of H. sendtneri from Ljubić and Pribinić (B&H). These scattered serpentine vary regarding the concentration of Fe and Ni. Sparse vegetation further makes Ljubić locality more exposed to high intensity of solar radiation. POX and phenolics, are important in plant adaptation response to increased concentrations of heavy metals. POX activity differed in leaf and rhizome at both locations. Hydroxybenzoic acid was the most abundant phenolic in the leaves and ferulic acid in the rhizomes at both locations. The content of HM in the leaves and rhizomes from both localities decrease in following order: Fe>Ni>Cu>Cr; Fe>Ni>Cr>Cu respectively. The rhizome is directly exposed to HM and the response to excess HM may include increased lignification, metal chelation, and antioxidants activation. We discuss the role of POX and phenolics in adaptation of H. sendtneri to HM excess.

Keywords: serpentinophyte, serpentine soils, antioxidant activity, copper, iron, nickel, chromium.

PUTNIK-DELIĆ, Marina

Growth and water regime of some Brassica species under the stress caused by nickel and cadmium

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Four species from Brassica genus, rapeseed (Brassica napus L.), white mustard (Brassica alba L.), black mustard (Brassica nigra L.) and turnip (Brassica rapa L.), were analyzed after growth in the continuous presence of 10-5 M Ni or Cd, since sowing, intending to assess effects of Ni and Cd on growth and plant water regime.
Before sowing, seeds were kept for 24 hours in deionized water (control), 10-5 M Ni (as NiSO4) or Cd (as CdCl2). Seeds were germinated in the quartz sand, in an incubator, at 26°C. Seedlings were planted in pots containing ½ strength Hoagland nutrient solution (control) to which were added Ni or Cd to a final concentration of 10-5 M. The experiment was set in 5 replications with 8 plants per replication for each plant species. The nutrient solution was changed every other day and aerated regularly. After 30 days of growing, plants were analyzed.
The leaf area of plants treated with heavy metals was significantly smaller in comparison to the control, especially in the presence of Cd. The ratio between shoot and root dry weight declined in the presence of both Ni and Cd, except in black mustard where the difference was statistically insignificant. This suggests that Ni and Cd affected stronger shoot than root biomass. An increase in transpiration intensity in the presence of Ni, concomitantly with the reduction in biomass production, suggests that the transpiration coefficient in those plants increased, which is highly undesirable.

PYPKA, Magdalena

Molecular response of tobacco plants to heterogenous Zn availability

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Zn is a key trace element for the growth and development of all living organisms and plants are the main source (directly or indirectly) of Zn in the diet. Zn distribution in soil is heterogeneous and it changes during growth season, reducing plant Zn uptake efficiency. Nevertheless, plants can adapt their growth to the signals coming from the environment (i.e. gravity, light, availability of water and nutrients). In the case of Zn, it was shown that plants can detect Zn patches in the soil and alter their root growth to optimize Zn uptake. The Zinc-tropism (a kind of chemotropism) is not well understood. However, the successful detangling of Zn-tropic mechanisms in plants may pave the way for the new Zn biofortification approaches, including increasing the efficiency of Zn fertilizers.
My research aims to understand root Zinc-tropism, its relation to plant Zn-status and Zn homeostasis. To do that I use tobacco (Nicotiana tabacum var Xanthi) plants grown in standard agar media and Transparent Soil (TS). TS is hydrogel-based 3D medium that mimic soil. It has modular grain size, access to the air pockets, holds water and could be an adjustable nutrient source. Using TS allows me to heterogeneously distribute Zn and track the growth of roots, identifying those that grow toward the Zn source. Using molecular methods I identify potential Zn-tropic genes that could play a role in Zn detection and mechanisms that allow the root tip to change its growth direction along the Zn gradient. The initial results of my experiments suggest that various Zn placements in the soil affect the growth of the roots and have a specific impact on the expression of Zn transporters (ZIPs).
The research is realized with the funds of the National Science Center as part of the SONATA project (2020/39/D/NZ9/02393).

RELIC, DUBRAVKA

Bioaccessibility of potentially toxic elements and health risk assessment from urban parks soil samples from Central Banat District in Serbia

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Children spend the majority of their time in parks and playgrounds in urban areas, yet the health risk posed by the urban pollutants that probably end up in these areas is significant and generally neglected. When young children play on the playground, they may ingest more potentially toxic elements (PTE) and other contaminants from the soil due to hand-to-mouth activity, especially since children's hands are frequently wet, and the adhesion of dirt particles increases and raises the amount of particles swallowed. There are numerous of in vitro oral bioaccessibility methodologies, some of them, like the Simple Bioaccessibility Extraction Test (SBET), merely replicate the gastric microenvironment. Considering only some of the solubilized element ions in stomach solutions are absorbed, this approach can only yield "conservative" estimation. Physiologically Based Extraction Tests such as Unified BARGE Method (UBM), a biomimicry mouth-gastric-intestinal approach, simulates the entire digestive system. In addition to utilizing the SBET and UBM extraction methods to assess the bioaccessibility fraction of PTE, soil samples from parks were microwave digested to determine the pseudo total concentration of elements, which is typically taken into account for health risk assessment. However, not all of the pseudo total fraction of PTE from soils is available for absorption in the body; the bioaccessibility fractions are more specifically for that. Sulfur was complexed with glycine and only extracted employing the SBET in vitro approach, while Mo was only present in the gastrointestinal fraction of UBM in addition to their pseudo-total content. The significant concentrations of As, Cd, Cr, Pb, Cu, V, and Zn were obtained under gastro in vitro extraction conditions (UBM) in examined soil samples, there are worries about the likelihood of adverse health effects on the children body when these PTE are extracted in the stomach and made available to the systemic circulation.

RENELLA, Giancarlo

Uptake of thallium and other potentially toxic elements in vegetables grown in a pyrite mining contaminated soil: risk assessment and mitigation by soil organic amendment

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Thallium (Tl) is a potentially toxic element (PTE) receiving increasing public and governmental concerns due to past and current emissions, and it is listed among the 13 priority PTEs by the U.S. Environmental Protection Agency, European Commission (EC) and World Health Organization (WHO) because it acts as a neurotoxin, and prolonged human exposure cause gastroenteritis and even death. Due to its toxicity, Tl concentration in soil is regulated, but while in unpolluted areas Tl concentration in soil depends on the contribution from the weathering processes of parent material during pedogenesis, in polluted environments Tl concentration can reach values far higher than the legislation thresholds. posing risks for human health when polluted soils are used for crop production. We studied the Tl uptake by horticultural plants with different metal uptake characteristics, in grown on a soil polluted by Tl due to dismissed mining activities, subjected to mineral fertilization or amendment with compost. A field cultivation trial of Tuscany kale and red chicory was conducted on a Tl-polluted soil in Valdicastello Carducci (Tuscany, Italy), and various geochemical and health risk indices of PTEs in soil and vegetables were calculated.

Results showed that the studied soil was polluted by Tl, but also by Pb, As, Cd, Sb, Zn, and contained high concentrations of Ba, due to the presence of barite (Table 1). Kale leaves accumulated high Tl concentrations whereas chicory did not, kale leaves also displayed high concentrations of other PTEs and Ba at levels higher than recommended in foodstuff. Compost amendment of soil significantly reduced the Tl uptake by kale plants, not of other PTEs, but calculation of oral reference doses (RfD) indicated risks due to Tl intake associated to kale consumption and to Cr for both kale and chicory. Perspectives for protection of residents' health and potential of organic amendments as environmental mitigation measures are illustrated and discussed.

SALINITRO, Mirko

Root growth response to localized selenium enrichment in soil: a comparison between hyperaccumulator and non-accumulator species.

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Plant root foraging is a process that consist of roots preferential proliferation towards the soil microenvironment in which the resources are more abundant for plant itself. This process has been documented for many hyperaccumulator species which have the ability to modify their root architecture in presence of the accumulated element. To date, data on Se-hyperaccumulators root foraging are still rare and mostly regards Neptunia amplexicaulis.

The aim of this study was to determine whether the Se-hyperaccumulator species Stanleya pinnata and Astragalus bisulcatus actively forage for selenate (SeO24-) or selenite (SeO23-) in soils, compared to the non-accumulator species Brassica juncea and Medicago sativa.

Plants were grown in rhizotrons half-filled with Se-enriched soil (in form of selenate or selenite, 30 mg/kg) and half-filled with control soil. Young seedlings were transferred at the interface between the two soils and grown for three weeks. Root development was evaluated through image analysis.
The Se-hyperaccumulator S. pinnata, when grown in presence of selenate, developed the 76 % of roots in the selenate-enriched half and the 24% of roots in the control half. No significant differences were detected for selenite treatment.
The Se-hyperaccumulator A. bisulcatus did not show any sign of root foraging either in selenite and selenate treatment, developing an equal density of root in Se-enriched soil and the control halves. The non-accumulator B. juncea, and M. sativa grown under the same condition did not show any preferential distribution of roots either for selenate or selenite treatments. In few rhizotrons the two species showed signs of Se-avoidance developing more roots in the control part compared to the Se-enriched one, despite these results were not statistically significant.

SARTHOU, Manon

The involvement of calcium homeostasis in uranium uptake.

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Uranium (U) is an element naturally occurring in the biosphere and taken up by plants. However, U is a non-essential element that is highly toxic for living organisms. In this study we looked at the effects of media nutrient deficiencies on the accumulation of U in Arabidopsis thaliana roots. We provided evidences that U accumulation was strongly related to macronutrient homeostasis, particularly to calcium (Ca) homeostasis, and was taken up through Ca2+-permeable cation channels. Firstly we showed that U accumulation was strongly increased under macronutrient deficiency and Ca deficiency was the only single cationic macronutrient deficiency able to induce an increase in U accumulation in Arabidopsis roots. Then we showed that exogenous Ca addition was able to reduce U accumulation and that the Ca2+-permeable channel inhibitors gadolinium, verapamil and nifedipine were able to significantly decrease U accumulation, suggesting that U could use the same route as Ca to enters the roots. Finally, we showed that the accumulation of U in the single mutants mca1 and ann1 impaired for the MCA1 and ANN1 Ca2+-permeable channels presented a reduction of U accumulation in roots of about 40 %. Taken together, our results show the importance of Ca homeostasis and Ca2+-permeable channels in U accumulation in A. thaliana roots.

SIEMIANOWSKI, Oskar

Zn-tropism: signal detection and control over the growth direction – the many ways for plants to explore and exploit environment

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Plants provide almost all the calories and microelements needed for the human diet. Plant productivity depends mostly on water access and nutritional uptake. Conventional approaches to increase plant nutritional value (biofortification) include increasing nutrient uptake, facilitating root-to-shoot nutrient translocation, and increasing relocation (transport) of microelements into the seed. Increasing the efficiency of plant roots to find micronutrients in soil could support those efforts! The phenomena of plant sensing and moving toward source of nutrients is called chemotropism (or nutritropism). The mechanism of sensing nutrients and control over the growth direction toward microelements is a subject of intense research but not many results. This is partially due to limited visibility of roots during growth or lack of control on the distribution of elements in the growth medium. I developed an affordable custom tools to overcome those issues: (i) a hydrogel-based transparent soil and (ii) a paper microfluidic growth platform. Both allow for in situ root tracking and provide spatial distribution of water/nutrients access points. Using those tools, I showed a unique mechanism of water potential independent hydrotropism of the whole root system of Brassica rapa. Using the same tools, we are currently investigating the potential mechanisms that would allow roots to turn toward Zn source (Zn-tropisms). Our focus is on the role of the cell wall and its flexibility that depends on the cation that crosslinks the homogalacturonan. Zn ions have more affinity and result in more rigid pectin gels. Our hypothesis is that the root Zn-tropism mechanisms, might result from the contact of one of the root sites with Zn, which would cause stronger pectin crosslinking, resulting in less flexible CW and decrease cell elongation. On the other root side, without Zn, cell wall would be relatively more flexible and therefore, cells would elongate, promoting root turning toward Zn.

SMOLDERS, Eric

Managing Cadmium in Agricultural Systems

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Cadmium (Cd) is a naturally occurring element present in all soils but its concentration has been increased by application, sewage sludge, animal manure and by atmospheric deposition. The primary risk pathway to humans is exposure through dietary intake in foods and by inhalation through smoking. Despite numerous regulations to limit Cd exposure, risks remain and may even increase in some developing countries. This paper will revise the main pathways of Cd in the journey of Cd between soil and the human gut with due attention to health risks and regulations worldwide. Our understanding of the mechanism of root Cd uptake, translocation and accumulation in shoots, grains and tubers has advanced significantly and farmers now have the option to manage Cd through choice of crop and/or cultivar. At the same time, genetic technologies offer great promise to develop low Cd-accumulating cultivars through our knowledge of the genes responsible for Cd uptake, translocation and sequestration in plants. High-risk soils can now be identified by soil testing and various agronomic or post-harvest options are available to minimise risks. Combined, these management options will have much quicker results in reducing Cd concentrations in foods than reductions in Cd inputs in fertilizers, manures or atmospheric sources.

SMOLEŃ, Sylwester

Possibility of using iodoquinolines to enrich of kale (Brassica oleracea L. var. sabellica) with iodine

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Two varieties of kale cv. 'Oldenbor F1' and 'Redbor F1' was grown in a hydroponic study in a greenhouse. The following treatments were the subject of the experiment: 1) Control; 2) quinoline /without iodine negative control for iodoquinolines/, 3) KIO3; 4) 8-Hydroxy-7-iodo-5-quinolinesulfonic acid; 5) 5-Chloro-7-iodo-8-quinolinol and 6) 5,7-dijodo-8-chinol. Their applications in 10 µM concentrations started at the stage of 4-5 true leaves and was continually until harvesting at phase of 13-14 true leaves. The tested compounds had no effect on plant biomass. The content of iodine in kale plants after the application of individual iodine compounds was as follows: 8-Hydroxy-7-iodo-5-quinolinesulfonic acid=KIO3>5,7-dijodo-8-chinol>5-Chloro-7-iodo-8-quinolinol>quinoline>control. The performed analyzes (including ICP-MS/MS, ICP-OES, LC-MS/MS, capillary electrophoresis techniques) showed different chemical composition of kale plants.
In summary: iodoquinolines were taken up by the roots and transported to kale leaves. These compounds can be used to enrich plants with iodine.
Acknowledgements: grant No.: UMO-2020/37/B/NZ9/02710 NCN, Poland. The use of individual iodoquinolines for enriching plants with iodine is proprietary by several patent applications in the Polish Patent Office.
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STRATULAT, Tatiana

Influence of Foliar Applications of Potassium Bicarbonate on Cucumbers on the Dynamics of Potassium Accumulation in Leaves

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Potassium (K) is one of the main plant nutrients and is involved in many important physiological processes. Young organs of plants contain K at levels 3-5 times higher than the old ones. With a sufficient content of K the resistance of plants to diseases increases. Scope of study: to evaluate the effect of foliar treatments of cucumbers against powdery mildew with aqueous solutions of potassium bicarbonate (KHCO3) on the levels of K content in the leaves of the plant. Six foliar applications were carried out during the period of vegetation with 0.3% and 0.4% aqueous solutions of the fungicide, containing 200 g KHCO3/kg. Sampling of cucumber leaves (10 leaves/dose, including control) occurred after 4th and 6th treatment; 3 weeks and 1.5 months after the last treatment. Leaves were dried in the air. Preparation of samples for the determination of K was carried out according to the accepted method, ashed at 450-500°C. The determination of K was carried out on an AAS-1 in the emission mode, in a propane-butane-air flame. The treatment of bushes with KHCO3 had a significant impact on the dynamics of K accumulation in cucumber leaves during and after treatment: K content in the control was at 18-20% lower than in the treated leaves, amounting to 8.583 g/kg at the end of the study, compared to 11.16 g /kg (0.3% KHCO3) and 10.509 g/kg (0.4% KHCO3), which extended the growing season by 6 weeks, increased the resistance of plants to powdery mildew and the keeping quality of fruits.

ŠINŽAR-SEKULIĆ, Jasmina

Ionomic characteristics of Dactylorhiza sambucina, a pseudo-metallophyte orchid species thriving in various soils in the Balkans

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Orchids have the remarkable ability to thrive in almost any terrestrial habitat, even in soils that may contain toxic metals. In Europe, the orchid species Dactylorhiza sambucina (L.) Soó is widely distributed and demonstrates its adaptability as a generalist, inhabiting different habitat types across various geological substrates. The aim of this study was to investigate whether the distinct geological substrates, particularly ultramafic ones characterized by unfavourable physico-chemical properties, affect the elementome profiles and ecophysiological responses of this orchid. To address this question, we analysed plants from eight populations of D. sambucina from three substrates types: ultramafic, calcareous, and siliceous. The differences in elementome profiles are discussed in terms of the predominant tendency in orchids to accumulate macroelements in aboveground plant organs, whereas trace elements tend to be excluded. The study of ecophysiological parameters, such as the concentrations of photosynthetic pigments and phenolic compounds, was carried out to additionally identify differences between populations from different geological substrates, with the lowest stress level expected in plants from calcareous soils.

ŠKONDRIĆ, Siniša

Serpentine flora of Bosnia and Herzegovina

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The Balkan Peninsula is considered one of the main biodiversity centers of Europe. Also, in Europe, the largest areas under serpentine substrate, as well as the main center of diversity of serpentine flora, are located in the Balkans. In Bosnia and Herzegovina, serpentine sites can be divided into four groups: 1) Prijedor-Banja Luka, 2) Borja and Ozren, 3) Žepče-Gostović-Krivaja and 4) Višegrad-Rudo. The serpentine soils are characterized by: 1) high levels of heavy metals: iron, nickel, chromium and cobalt, 2) nutrient deficiency, especially in nitrogen, phosphorus and potassium, 3) low calcium-magnesium quotients and calcium deficiency and 4) lower levels of clay colloids and clay minerals with a low exchange capacity. The specific physical and chemical properties of the serpentine substratum caused the existence of a significant number of local, regional and transregional endemics in the Balkans. Three main groups of Balkan endemics can be recognized according to their preference for the substrate: obligate serpentine endemics, facultative serpentine endemics and accidental serpentine endemics. In Bosnia and Herzegovina, most Balkan endemics of serpentine flora are listed on the Rulebook of strictly protected and protected wild species in the Republic of Srpska. The work presents data that contribute to a better knowledge of the serpentine flora of Bosnia and Herzegovina.
Key words: serpentinophytes, Balkan, endemism

THOMINE, Sebastien

Regulation of intracellular free zinc concentration in Arabidopsis thaliana roots

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Zinc (Zn) is an essential micronutrient for all living organisms. Zn malnutrition affects one third of the global human population and plants are the main entry point for Zn into the food chain (Assunção et al., 2022, 10.1093/jxb/erac014). Enhancing Zn accumulation in crops is required to improve plant nutritional value and growth in nutrient-deficient soils. Zn uptake in plant root is regulated by F-bZIP transcription factors that directly perceive intracellular Zn concentrations ([Zn]i; Lilay et al., 2021, 10.1038/s41477-021-00856-7). Here, we use eCALWY, a genetically encoded fluorescent Zn sensor, expressed in the cytosol of Arabidopsis cells to investigate the regulation of [Zn]i in root cells (Lanquar et al. 2014, 10.1111/nph.12652). [Zn]i lies in the 100 pM range and increases transiently upon Zn elevation in the medium. We investigate [Zn]i in mutants lacking Zn transporters. This research will highlight the mechanisms leading to Zn deficiency responses or toxicity.

TORRE, Sissel

Accumulation of Manganese and Zink causes outer leaf marginal necrosis in lettuce

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X-ray Fluorescence Techniques (XRF) were used to image the composition and distribution of ions in healthy and injured outer tips of lettuce. High accumulation of Mn, Zn and Ca, and depletion of K and Cl was observed in injured parts of the tips exposed to high irradiance. Very early (small necrotic spots), corresponding to the area with high stomatal density, showed high accumulation of Mn and Zn. The striking changes in accumulation and distribution of Mn and Zn between healthy and injured parts suggest local metal toxicity to play a role in inducing outer leaf marginal necrosis in lettuce. Accumulation of reactive oxygen species (ROS) was detected in outer tips and callose deposition was identified in the border between living and dead cells in injured tips indicating that increased light intensity triggers a defensive metal response in the tips of outer mature leaves.

VACULIK, Marek

The effect of silicon on uptake and physiological process in plants exposed to arsenic and antimony

Marek Vaculik[1]; Miroslava Vaculikova[1]; Adriana Misuthova[2]; Rajpal Shetty[2]; Nair CS Vidya[3]; Zuzana Lukacova[2]; Boris Bokor[2]; Alexander Lux[2]; Ivana Fialova[3]; Miroslava Luxova[3]
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Silicon (Si) is the 2nd most abundant element in the Earth crust and considered as beneficial element in plant nutrition. Although Si is not considered as an essential element for plants in general, many plant species could take up large amount of this metalloid that exceed those of other essential macronutrients like phosphorus (P) or sulphur (S). Moreover, Si was found to ameliorate various forms of abiotic and biotic stresses. In the last decades, several reports documented that phytotoxicity effects induced by excess metals like Cd, Zn or Pb, as well as metatalloids like As and Sb could be mitigated by this beneficial element. The Si-based mechanisms allowing plants to cope with this type of abiotic stress has been developed at multiple levels in various plant species. Our research on various plant species, including maize, sorghum, Arundo donax, Nicotiana benthamiana and some other species showed that Si could affect the uptake of As and Sb by influencing the development of root apoplasmic barriers, and influencing the genes responsible for Si uptake. Additionally, several biochemical and physiological processes, like the activity of antioxidant enzymes, stability of cell membranes, photosynthesis and production of lignin are affected in those plants grown in the simultaneous presence of Si and toxic metalloids like As and Sb. This work was supported by the Slovak Research and Development Agency (grant number APVV-17-0164 and APVV SK-CN-21-0034), and by Slovak Grant Agency VEGA (grant number VEGA 1/0472/22, VEGA 2/0103/21), and by a COST Action Nr. COST CA19116.

VAN DER ENT, Antony

X-ray fluorescence microscopy for unravelling the secrets of hyperaccumulator plants

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Hyperaccumulators are attractive models for studying metal(loid) homeostasis and probing the spatial distribution and coordination chemistry of metal(loid)s in their cells and tissues is important for advancing our understanding of their evolution, ecophysiology, biochemistry, and molecular biology. Methods based on the X-ray fluorescence (XRF) phenomenon are attractive because they are intrinsically non-destructive and give access to most elements in the periodic table. Synchrotron X-ray fluorescence microscopy (XFM) is a powerful technique based on XRF to produce elemental maps of samples by raster-scanning the sample through a focused X-ray beam and collecting and analysing the emitted XRF photons in each pixel. XFM can be used to quantitatively determine elemental concentrations in plant samples attaining a spatial resolution of 0.5–2 µm and detection limits down to 10 µg g-1 for transition element in areas of up to 100x100 mm in size. This approach is unique in providing in situ information, and with appropriate sample preparation offer results true to biological conditions of the living plant. New fast detector systems enable mapping of not only hydrated plant organs, such as leaves, but of whole live plants and even of time-series after exposure to a metal(loid). The last development in this field is the practical implementation of XFM computed tomography (XFM–CT) which enables “virtual sectioning” of a sample thereby entirely avoiding artefacts arising from destructive sample preparation. XFM can play a key role in answering questions at every level of metal(loid) homeostasis and regulation in plants, from the rhizosphere interface to uptake pathways in the roots and translocation through the shoot. Further improvements in the technical capabilities of synchrotron facilities and laboratory-based systems offer exciting perspectives for the investigation of hyperaccumulator plants into the future. In this presentation I will show various examples of hyperaccumulator studies to illustrate the usefulness of this technique.

VERBRUGGEN, Nathalie

Cadmium tolerance and detoxification in plants: what can we learn from Arabidopsis halleri?

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Cadmium is one of the most toxic trace metals for living organisms and the highest concentrations have been found in plant cells. Cadmium accumulation in the environment is recognized as a worldwide concern. The accumulation pathway of cadmium in plants is crucial for phytoremediation applications, but also in breeding programmes to limit its consumption by crops. However, the different mechanisms used by Cd to enter, be transported and detoxified in plants remain unclear. In this presentation, the contributions made through the studies on Arabidopsis halleri will be outlined.

Arabidopsis halleri is a diploid perennial , self incompatible, and close relative to the plant model Arabidopsis thaliana. Arabidopsis halleri is considered as a model species for the study of metal homeostasis and detoxification. The species seems to constitutively hyperaccumulate zinc while cadmium accumulation is more variable. Some populations do however accumulate cadmium while others accumulate much less than A. thaliana.

A.halleri is a pseudometallophyte, able to grow on metal contaminated or non-contaminated soils. In Europe the species is distributed in several genetic units within which metallicolous populations seem to have been established from metallicolous populations. The comparative study of populations showed different mechanisms of cadmium detoxification but a genetic determinant of cadmium tolerance and accumulation that looks common

In the presentation, contributions made to the understanding of cadmium accumulation and detoxification through studies on Arabidospis halleri will be outlined.

VISIOLI, Giovanna

Heavy metals modulate DNA compaction and methylation at CpG sites in the metal hyperaccumulator Arabidopsis helleri

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In this study we assume that at subcellular level, different strategies were adopted by hyperaccumulator vs the non-accumulator plant species to face the excess of heavy metals. At this purpose the comet assay was used to investigate the nucleoid structure modifications occurring in response to Zn and Cd treatments in the I16 and PL22 populations of the hyperaccumulator Arabidopsis halleri vs the non-accumulator species Arabidopsis thaliana. Methy-sens comet assay and RT-qPCR were also performed to associate metal induced variations in nucleoids with possible epigenetic modifications. The comet comet assay showed that Zn induce a mild but non-significant reduction in the tail moment in A. thaliana and in both I16 and PL22. Cd treatment induced an increase in DNA migration in nuclei of A. thaliana, whereas no differences in DNA migration was observed for I16, and a significant increase in nucleoid condensation was found in PL22 Cd treated samples. This last population showed higher CpG DNA methylation upon Cd treatment than in control conditions, and an up-regulation of genes involved in symmetric methylation and histone deacetylation. Our data support the hypothesis of a possible role of epigenetic modifications in the hyperaccumulation trait to cope with the high Cd shoot concentrations. In addition, the differences observed between PL22 and I16 suggest divergent strategies for metals detoxification developing in the two metallicolous populations.

VÍTOVÁ, Milada

Accumulation of rare earth elements from waste of CFL bulbs by the red alga Galdieria sulphuraria

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High cumulation of compact fluorescent lamp (CFL) waste on the one hand, and the criticality of the rare earth elements (REEs) supply for electronic devices and modern technologies on the other hand leads to the need of their use for recovery. The widely used CFL lights, and their wastes are good secondary sources of REEs. Bio-removal of REEs would be an environmental friendly option of the recovery. To address this problem, the current study focusses on the use of the extremophilic red alga, Galdieria sulphuraria, for bioaccumulation/removal of REEs from CFL acid extract, and the physiological response of a synchronized culture of G. sulphuraria. A CFL acid extract significantly affected growth, photosynthetic pigments, photosynthetic activity, and cell cycle progression of this alga. The synchronous culture was able to efficiently accumulate REEs from a CFL acid extract and efficiency was doubled by including two phytohormones i.e., 6-Benzylaminopurine (BAP - Cytokinin family) and 1-Naphthaleneacetic acid (NAA - Auxin family). Accumulation of REEs by G. sulphuraria was concentration-independent and selective.

WIENKOOP, Stefanie

Role of Ferritin(s) in nodule formation and the symbiont induced stay-green effect upon drought

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The mutualistic relationship between nitrogen-fixing rhizobacteria and legumes play a crucial role in enhancing the plants tolerance to drought stress. Previous research has demonstrated that this symbiosis triggers a phenotype known as symbiont induced stay-green (SISG), which delays leaf senescence in response to drought and facilitates quicker recovery from desiccation. Ferritin, a Fe-storage and distribution protein, known to be involved in stay-green upon drought stress, seemed to play a major role in this phenotype. Their levels were enhanced during symbiosis compared to non-symbiotic Medicago truncatula (Jemalong) plants.
Here, we show that Ferritins are additionally involved in nodule formation. Knockdown mutants show reduced nodulation while function was retained. Hence, Ferritin supports the formation of the symbiotic relationship and symbiosis, vice versa, induces the levels of Ferritin. As a consequence, ferritins are not only regulated by rhizobia but also regulating symbiosis. In order to decipher the dynamics of the different isoforms of Ferritin in this relation(s), we further analyzed the absolute amount of ferritin isoforms in leaves of M. truncatula wildtype R108. For this, we were using the Mass Western approach and compared Ferritin levels upon symbiosis and drought. The results will be discussed in this poster.

WIGGENHAUSER, Mattthias

The potential of Swiss and EU wheat cultivars to close trace metal gaps in plant-based diets

Mattthias Wiggenhauser; Lisa Konrad; Lorenz Luder; Andreas Hund; Julie Tolu; Lenny Winkel; Lukas Kronenberg; Isabelle Herter-Aeberli
ETH Zürich - Switzerland (CH)

The FAO expects a shift towards plant-based diets in wealthy countries such as Switzerland due to environmental, health, and animal welfare concerns. This shift can critically reduce the intake of trace metals such as zinc (Zn), iron (Fe), and selenium (Se), particularly in vegan diets. As wheat could be a potential source to close this 'micronutrient gap', we measured for the first time the trace metal concentrations in Swiss wheat elite genotypes that grew on a weakly alkaline soil in the years 2016, 2017, and 2019. We further determined the concentrations of phytate that decrease the bioavailability of Zn and Fe for humans. The concentrations in the wheat grains ranged from 16 to 28 mg kg-1 for Zn and 25 to 42 mg kg-1 for Fe between the different genotypes. Compared to other countries, these concentrations are neither high nor low. Analyses of variance revealed that the genotypes and the year of sampling had significant effects on these concentrations. Phytate to trace metal molar ratios were high for Zn (> 15) and Fe (> 1), indicating a low bioavailability. In contrast to Zn and Fe, neither the cultivar nor the year of sampling determined the Se concentrations in the grains, but the location within the field. This preliminary data set suggests that i) annual meteorological conditions and genotype can have a strong impact on Fe and Zn concentrations in wheat grains, ii) Se concentrations may depend more strongly on soil conditions than on wheat genotypes, and iii) there is potential to increase the bioavailability of trace metals in Swiss wheat through agronomic and genetic biofortification. At the conference, we plant to additionally present data of EU wheat genotypes (n = 80) and food-basket based calculations to estimate if biofortified wheat could significantly improve trace metal intake in plant-based diets.

YASUOR, Hagai

Involvement of trace metal nutrition in plant physiology process and abiotic stress responses

Hagai Yasuor
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Trace metals have a crucial role in different plant physiological processes. Low concentrations of these elements are needed for normal plant growth and development. It also plays a role in plant responses to biotic and abiotic stresses. Our research focuses on the effect of metal nutrition on reproductive physiology (flower and fruit organ development and function) and fruit quality, mainly understanding the role in mitigating abiotic stress adverse effects. For example, zinc imbalances affect maize male inflorescence development, manganese enrichment effects on pepper heat damage, and the possible role of calcium in these physiological disorders. Also, we are interested in understanding sulfur deficiencies in iron-sulfur complexes' activity in nitrogen fixation processes in legume crops (peanuts). The sulfur deficiencies might be relevant due to future sulfur deficiencies in agricultural areas worldwide.

XUE, Peiying

Foliar uptake of Pb from atmospheric deposition is a major source of Pb accumulation in wheat grain

Peiying Xue[1]; Wenju Liu; Liuxu Feng; Peipei Gao
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Atmospheric heavy metal deposition in agroecosystems has increased recently, which poses serious risks to crop safety and human health via food chain. According to our previous research, wheat grains can accumulate high levels of Pb even when wheat is planted in soils with low levels of Pb. The Pb isotopic data of a field research showed that airborne Pb was the major source (> 50%) of Pb in wheat grain. Furthermore, when wheat flag leaves were exposed to PbSO4 at the booting stage, and much greater Pb accumulation (0.33-0.48 mg/kg) was observed in exposed wheat grain than in the control ($P < 0.05$), PbSO4 constituted most (82.80-100%) of the Pb in the wheat grain. These results confirmed the efficient foliar Pb uptake and transfer from atmospheric deposition into wheat grain.

Field-culture experiments with 57 winter wheat cultivars were conducted at two Pb contaminated sites in North China to screen low Pb accumulated cultivars. They showed significant difference in Pb accumulation in wheat grain, ranging from 0.55-0.07 mg/kg (Site 1), 0.40-0.07 mg/kg (Site 2), which indicated that selection of wheat cultivars accumulating low levels of Pb could provide an efficient path to compliant with the grain Pb limits of the Chinese Food Safety Standards (0.2 mg/kg, GB2762-2017). In addition, 25 wheat cultivars were selected to further investigate their root and leaf Pb uptake abilities. The high differences of Pb accumulation in wheat grain mainly resulted from different ability of foliar uptake of Pb rather than that of root uptake. Leaf Pb concentration correlated positively with stomatal width and trichome length. A hydroponic experiment also conducted to prove that the open stoma is an important pathway of foliar particulate Pb into spikes of wheat even after short-term exposure for 6 h, contributing 33%-46%. This would be a new sight for understanding the contribution of airborne Pb to Pb accumulation in wheat grains.

ZHAO, Fang-Jie

Cadmium biogeochemistry in paddy soils and strategies to reduce Cd accumulation in rice

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Rice is a staple food for about half of the global population, for whom rice is also a major dietary source of the toxic trace metal cadmium (Cd). Contamination of paddy soils with Cd is a common problem in some areas of Asian countries, resulting in elevated intake of Cd and significant health risk for the local residents. It is therefore imperative to develop strategies to limit Cd accumulation in rice. The concentration of Cd in rice grain varies widely, from below the detection limits to several mg/kg. This wide variation is caused by the variations in the level of Cd contamination in the soil, soil properties and biogeochemical processes that affect Cd availability to plants, and the genetics that govern the uptake and distribution of Cd in rice plants. Paddy soil undergoes episodic flooding-draining cycles during the rice growing season, leading to wide fluctuations in the redox potential, a key factor controlling Cd availability in the soil. Cadmium availability decreases rapidly under anoxic conditions due the formation of insoluble cadmium sulphide; this process is reversed when paddy water is drained. The rate of cadmium remobilization after drainage of paddy water is controlled by factors such as the voltaic cell effect, production of free radicals, and pH-dependent sorption processes. These processes can be controlled to slow down the remobilization of Cd after drainage. There are large variations in Cd accumulation among different rice cultivars; the genetics underlying some of these variations have been elucidated in recent years. Several membrane transporters play important roles in the uptake, sequestration and distribution of Cd in rice plants, which are targets for manipulations to limit Cd accumulation in the grain. This knowledge has been used to enable breeding of ultra-low Cd accumulating rice cultivars.

ZHAO, Siting

Characterization of Arabidopsis Zinc Sensor Mutants

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Zinc-deficient soils are widespread globally and human Zn malnutrition affects about one-third of the world's population with a negative impact on growth, immune system and cognitive functioning, being more prevalent in populations that rely on cereal grains as staple food. Fundamental knowledge on the Zn deficiency response and Zn homeostasis regulation in plant crop species will contribute to developing crops with improved Zn nutritional content (biofortified) and improved resilience to Zn deficient soils.
Arabidopsis F-bZIP transcription factors, bZIP19 and bZIP23, play a central role in the transcriptional regulation of the Zn deficiency response. Under Zn deficiency, they bind to Zinc Deficiency Response Elements (ZDRE) in the promoters of their target genes that are transcriptionally activated. Target genes include Zn homeostasis genes involved in Zn transport and distribution. In addition to being the regulators of the Zn deficiency response, Arabidopsis bZIP19 and bZIP23 also function as Zn sensors, through a direct binding of Zn2+ ions to their Zn-sensor motif (ZSM), i.e. a cysteine/histidine-rich motif at the protein N terminus. Under Zn sufficient conditions, the Zn binding to the ZSM halts the transcriptional activity of bZIP19 and bZIP23. Deletion or amino acid substitutions in the ZSM disrupt the Zn-sensor function of bZIP19/23, which leads to a Zn-insensitive and constitutive transcriptional activation of the bZIP19/23 target genes. Analysis of Arabidopsis mutant lines expressing bZIP19 with a deleted ZSM showed a significant increase in plant and seed Zn accumulation. Here, we investigate Arabidopsis mutant lines expressing bZIP19 with amino acid substitutions in the ZSM. We investigate the effect of these ZSM mutations in plants grown with different levels of Zn supply, and analyse Zn accumulation, ionomics and selected genes expression.

ZHENG, HongXiang

Plasma Membrane-localized Transporter NREET1 is Responsible for Rare Earth Element Uptake in Hyperaccumulator Dicranopteris linearis

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Rare earth elements (REEs) are critical for an increasing number of technological applications, including clean energy, electric vehicles and electronic devices. However, REE mineral reserves are depleting, recycling yields are extremely low, and the production steps are resource intensive with a huge carbon footprint. Some plant species are able to hyperaccumulate REEs, and their biology could play a pivotal role in developing more environmentally-friendly REE recovery technologies. The mechanisms by which plants take up and control REEs remain poorly understood. To develop plant-based REE recovery technologies, we identified a REE-specific transporter NREET1 (NRAMP Rare Earth Element Transporter 1) from the REE hyperaccumulator fern Dicranopteris linearis. Although NREET1 belongs to the NRAMP (natural resistance-associated macrophage protein) family, it shares a low similarity with other NRAMP members. When expressed in yeast, NREET1 exhibited REE transport capacity, but cannot transport divalent metal such as zinc, nickel, manganese or iron. Expression studies in yeast and Arabidopsis thaliana revealed that NREET1 is localized predominantly in the plasma membrane of roots. Compared to the wild-type species, heterologous expression of NREET1 in Arabidopsis resulted in increased REE uptake and transfer from root cell walls into the cytoplasm. Moreover, NREET1 has a higher affinity for transporting light REEs compared to heavy REEs, which is consistent to the preferential enrichment of light REEs in field-growing D. linearis. We therefore conclude that NREET1 is a plasma membrane-localized REE transporter, which may play an important role in the uptake and consequently hyperaccumulation of REEs in D. linearis. This is the first REE transporter reported in plants, opening a new perspective for understanding the REE uptake mechanism and laying the foundation to better understand the biological effects of REEs in plants. Moreover, heterologous expression of NREET1 enhanced the REE accumulation in the model plant, demonstrating a potential application of synthetically engineered plants for recovering valuable REEs from the environment.

Editorial

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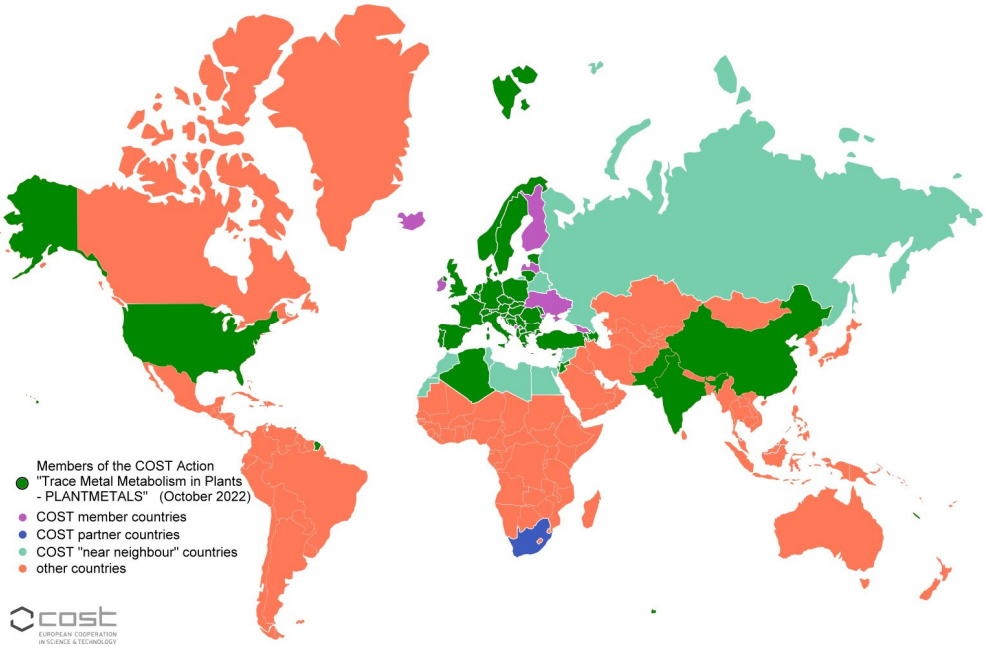

map showing the participating countries in this COST Action

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