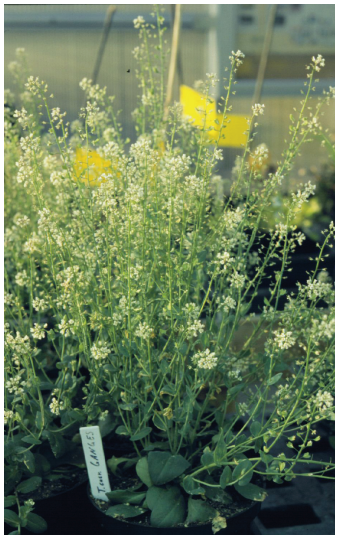
# Trace metal metabolism in plants

# PLANTMETALS Kick-off meeting

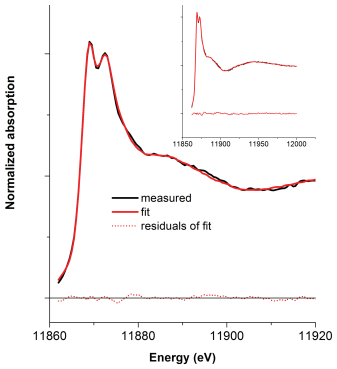
CA19116 Plant Metals

CA19116 Plant Metals

## - aimed at bringing together all members of the COST Action CA 19116



**24 - 27 August 2021**
**Location: Biology centre of the Czech Academy of Sciences, Main lecture hall and campus**

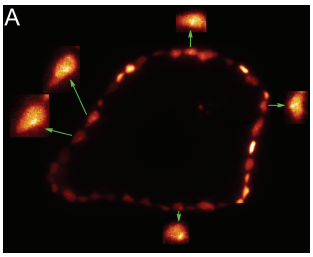



Programme:

**24 August**

15:00 to 17:00: Registration

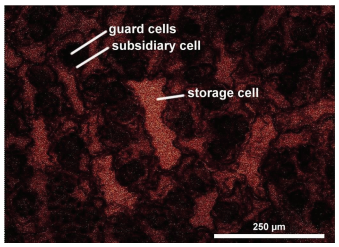
18:00-23:00: "Get together"- party (free for all registered participants)

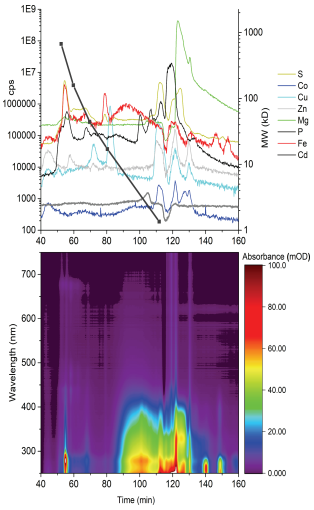



**25 August**

8:30-18:15 Lectures with 2x30min coffee breaks and 2h lunch break

*- free evening -*




**26 August**

8:30-12:00: Poster viewing with coffee

12:30-14:30: Lunch break

14:30-18:00: Management committee meeting (MC2)

19:00-23:00 Conference dinner in town

**27 August**

8:30-12:30: Workgroup planning

13:00 Lunch

cost
EUROPEAN COOPERATION IN SCIENCE & TECHNOLOGY

# Metals, Plants and People

# Contents

# Detailed programme

**IMPORTANT: COVID-19 hygiene according to current Czech rules:**
**- to enter a public inside room (except restaurant or other dining area), you need to wear an FFP2 mask.**
**- to enter an inside restaurant (for us, this also means the area of the coffee breaks incl. poster session), where naturally you cannot wear a mask while eating/drinking, you either need to show a certificate (ideally the EU digital certificate) of having been fully vaccinated with an EU-approved vaccine (ideally with the EU digital certificate). OR you need a certificate showing that you recovered from a COVID-19 infection not longer than 180 days ago. OR you need a certificate (ideally the EU digital certificate) of a test showing that you are currently not infected: PCR-test <1week or antigen test <72h.**
**- In the BC canteen where we have the lunches, additionally you need to wear the FFP2 mask except of the time where you sit down and eat/drink.**
**--> If you are NOT fully vaccinated or do NOT have the required certificate, get a PCR test before coming here to avoid any problems. Anyhow many airlines require that for flying.**

## 24 August, 2021: Welcome

15:00 – 17:00 Registration, hanging up posters in the P building of the Faculty of Arts of the University of South Bohemia – see the map at the end of this programme for the exact location.

18:00 – 23:00 "Get together"- party at the BCAS/USB Pavillion (“Klub Kampa”, https://cs-cz.facebook.com/KampaJU/), with buffet, grill and drinks
- see the map at the end of this programme for the exact location

– meant to bring all members of the COST Action together in an informal way, free for all registered participants (co-financing from KOROLID for those not covered by COST)

– the barbecue and buffet will be sponsored by COST, drinks are self-paid

– In good weather, the event is meant to be mostly outside, in bad weather inside the klub.
**IMPORTANT: COVID-19 hygiene: Please note that according to current Czech rules, to enter an inside restaurant you either need to show certificate (ideally the EU digital certificate) of having been fully vaccinated with an EU-approved vaccine (ideally with the EU digital certificate), or a certificate showing that you recovered from a COVID-19 infection not longer than 180 days ago, or a certificate (ideally the EU digital certificate) of a test showing that you are currently not infected by PCR-test <1week or antigen test <72h.**

## 25 August: Talks – meant to introduce the research of the WG leaders and main organisers and their vision of this COST Action

The talks have to be MS Powerpoint presentations, and each speaker has to send the final version of the talk to Robert.dulfer@umbr.cas.cz and Hendrik.Kuepper@umbr.cas.cz at least the day before, AND bring it to the lecture room on a USB stick. As we will stream the talks via Zoom to all off-site members of the Action, we need to use our PC, the use of own computers is not possible.

| Time | Speaker, affiliation | Title |
|---|---|---|
| 8:30-8:45 | Prof. Libor Grubhoffer<br>(director of the Biology Centre of the Czech Academy of Sciences = BCAS, České Budějovice, Czech Republic) <GH legal representative> | Welcome |
| 8:45-9:15 | Prof. Hendrik Küpper<br>(BCAS, IPMB, Dept. of Plant Biophysics and Biochemistry, Czech Republic) <chair, STSM coordinator, GH scientific representative> | Introduction to the COST Action "Trace metal metabolism in plants – PLANTMETALS"<br>and the scientific contribution of the workgroup of its chair |
| 9:15-9:45 | Prof. Nathalie Verbruggen<br>(Université Libre de Bruxelles, Faculté des Sciences, Laboratoire de Physiologie et de Génétique moléculaire des Plantes, Belgium)<br><vice chair, ITC conference grant coordinator> | Different strategies of Cd resistance in Arabidopsis halleri |
| 9:45-10:15 | **coffee break** | |
| 10:15-10:45 | Prof. Sebastien Thomine<br>(Institute for Integrative Biology of the Cell, Université Paris-Saclay, CEA, CNRS, France) <WG1 leader><br>Prof. Ana Assuncao<br>(Department of Plant and Environmental Sciences, University of Copenhagen, Denmark) <WG1 deputy leader> | A European perspective on the improvement of plant mineral micronutrient homeostasis for agriculture and human nutrition |
| 10:45-11:00 | Catherine Curie<br>(BPMP Montpellier, France)<br><WG1 talk selected from abstracts> | Manganese transport and its regulation in Arabidopsis |
| 11:00-11:15 | Ana Mijovilovich<br>(BCAS, IPMB, Dept. of Plant Biophysics and Biochemistry, Czech Republic)<br><WG1 talk selected from abstracts> | What X-rays can tell about metals in plants: localization and chemistry. Current and future work. |
| 11:15-11:45 | Prof. Eva Freisinger<br>(University of Zurich, Department of Chemistry) <WG2 leader><br>Prof. Moshe Sagi<br>(Ben-Gurion University of the Negev, Beer Sheva, Israel) <WG2 deputy leader> | WG2: Metalloproteins and -enzymes |
| 11:45-12:00 | Dr. Nadica Maltar Strmečki<br>(Ruđer Bošković Institute, Zagreb, Croatia)<br><WG2 talk selected from abstracts> | EPR spectroscopy as a tool to Investigate PLANTMETALS |
| 12:00-12:15 | Dr. Marie-Theres Hauser<br>(University of Natural Resources and Life Sciences, Vienna, Austria)<br><WG2 talk selected from abstracts> | Identification and characterization of a novel cell wall associated metal-binding peptide |
| 12:15-14:00 | ***lunch break – lunch served in canteen 13:00-13:30*** | |

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|---|---|---|
| 14:00-14:30 | Dr. Manuel Gonzalez-Guerrero<br>Centro de Biotecnología y Genómica de Plantas, Universidad Politécnica de Madrid, Spain)<br><WG3 leader><br>Dr. Marek Vaculik<br>(Plant Science and Biodiversity Centre of Slovak Academy of Sciences, Bratislava, Slovakia)<br><WG3 deputy leader> | Responses of plant Trace Metals to the environment |
| 14:30-14:45 | Tomica Mišljenović<br>(University of Belgrade, Faculty of Biology, Serbia)<br><WG3 talk selected from abstracts> | Bioaccumulation of potentially toxic elements in Noccaea kovatsii and N. praecox (Brassicaceae) from different geological substrates and their physiological responses to Ni |
| 14:45-15:00 | Dr. Filis Morina<br>(BCAS, IPMB, Dept. of Plant Biophysics and Biochemistry, Czech Republic)<br><WG3 talk selected from abstracts> | Beneficial role of zinc against biotic stress in metal non-hyperaccumulating plants |
| 15:00-15:15 | Dr. Irena Macek<br>(University of Ljubljana, Biotechnical Faculty. Slovenia)<br><WG3 talk selected from abstracts> | Long-term experiments in research of arbuscular mycorrhizal fungal community ecology |
| 15:15-15:45 | ***coffee break*** | |
| 15:45-16:15 | Prof. Philip White<br>(The James Hutton Institute, Dundee, Scotland, UK)<br><WG4 leader><br>Dr. Valerie Bert<br>(INERIS [Unité Technologies et Procédés Propres et Durables], France), <WG4 deputy leader> | Agronomic Aspects of Trace Metal Homeostasis |
| 16:15--16:30 | Hester Blommaert<br>Université Grenoble Alpes, France<br><WG4 talk selected from abstracts> | The Odyssey of Cd in Cacao Trees: from Soil to Bean |
| 16:30-16:45 | Dr. Sylwester Smoleń<br>(Faculty of Biotechnology and Horticulture, University of Agriculture in Krakow, Poland)<br><WG4 talk selected from abstracts> | Anticancer potential of lettuce biofortified with iodine in human gastrointestinal cancer cell lines as well as aspects of plant-derived thyroid hormone analogs in lettuce |
| 16:45-17:00 | Prof. Abdel Rahman Al Tawaha<br>(Al Hussein Bin Talal University, Jordan)<br><WG4 talk selected from abstracts> | Effects of foliar application of boron (B) on growth, yield and yield components and mineral composition of fenugreek |
| 17:00-17:30 | Prof. Ute Kraemer<br>(Ruhr-Universität Bochum, Germany)<br><WG5 leader, Science communication officer><br>Prof. Alexander Lux<br>(Comenius University in Bratislava, Faculty of Natural Sciences, Slovakia)<br><WG5 deputy leader> | Checkpoints of transport processes in plants facing toxic metals/metalloids and checkpoints of COST Action "PLANTMETALS" - CA19116 dissemination. |
| 17:30-17:45 | Dr. Muhammad Imran<br>(Nouryon, Amsterdam, Netherlands)<br><WG5 talk selected from abstracts> | Chelated micronutrient usage for overcoming crop deficiencies <dissemination: application of research findings to the field> |
| 17:45-18:15 | Dr. Martin Trtilek<br>(Photon Systems Instruments, Drasov, Czech Republic), <WG6 leader><br>Prof. Mark Aarts<br>(Wageningen Universiteit & Research, Netherlands)<br><WG6 deputy leader> | Intellectual Property rights in scientific project: visions and reality |

## 26 August Posters and MC meeting

| Time | Target group | Activity |
|---|---|---|
| 8:30-12:30 | **ALL members and supporters** | **Poster viewing with coffee and snacks – meant to show research and visions for the COST Action from ALL participants, incl. the speakers** |
| 12:30-14:30 | **ALL members and supporters** | ***lunch break – lunch served in canteen 13:00-13:30*** *afterwards: free afternoon for all non-MC participants* |
| 14:30-16:00 | **MC members / substitutes** | **2nd meeting of the management committee (1): Report of activities by the leaders of the Action: chair / STSM coordinator, vice chair / ITC conference grant coordinator, science communications officer, WG leaders incl. deputies** |
| 16:00-16:30 | **MC members / substitutes** | ***coffee break*** |
| 16:30-18:00 | **MC members / substitutes** | **2nd meeting of the management committee (2): Discussion and decision of goals and budget of the second grant period** |
| 19:00-23:00 | | ***Conference dinner in town*** Restaurant Hoch Spaliček, https://www.hochspalicek.cz/restaurace Cold+warm buffet ("all you can eat") sponsored by COST / organiser, drinks paid extra. **IMPORTANT: COVID-19 hygiene: Please note that according to current Czech rules, to enter an inside restaurant you either need to show certificate (ideally the EU digital certificate) of having been fully vaccinated with an EU-approved vaccine (ideally with the EU digital certificate), or a certificate showing that you recovered from a COVID-19 infection not longer than 180 days ago, or a certificate (ideally the EU digital certificate) of a test showing that you are currently not infected by PCR-test <1week or antigen test <72h** |

## 27 August- Planning of future workgroup activities

| Time | Target group | Activity |
|---|---|---|
| 8:30-9:00 | **ALL members and supporters interested in this WG**, others can view posters / meet individually | **WG1 – Metal transport: Activity planning for GP2**<br>*MoU description:* Pathways of TMs from the soil and through the plant. WG1 will identify the limiting factors in the uptake of plant physiologically active TMs from the rhizosphere and limit the uptake or the transfer to edible parts of toxic non-essential TMs WG1 will determine the chemical form (speciation) of TMs in each compartment. WG 1 will study crop plants and the use of hyperaccumulators and identify key nodes to improve/limit the uptake and accumulation of essential/non-essential TMs. WG1 will study interactions between TMs (antagonistic or synergistic effects).<br>Leader: Prof. Sebastien Thomine <Sebastien.THOMINE@i2bc.paris-saclay.fr>,<br>deputy leader: Prof. Ana Assuncao <agla@plen.ku.dk> |
| 9:00-9:30 | **ALL members and supporters interested in this WG**, others can view posters / meet individually | **WG2 – Metalloproteins: Activity planning for GP2**<br>*MoU description:* Metalloproteins important for TM homeostasis, metalloenzymes. WG2 will identify metalloproteins important for TM use efficiency. WG2 will study interaction of the proteins (other than transporters) with a TM: substrate affinity, regulation of activity, mechanism of function.<br>Leader: Prof. Eva Freisinger <freisinger@chem.uzh.ch>,<br>deputy leader: Prof. Moshe Sagi <gizi@bgu.ac.il> |
| 9:30-10:00 | **ALL members and supporters interested in this WG**, others can view posters / meet individually | **WG3 – Environment: Activity planning for GP2**<br>*MoU description:* Responses of plant TM metabolism to the environment. TM deficiency, TM toxicity, interactions with beneficial microorganisms and pathogens. WG3 will identify targets of both stress conditions and mechanisms, from the initial target to the whole- plant response. The initial targets can be lack or non-functional replacement of metal centres in metalloproteins, but also direct or indirect interactions with nucleic acids and metabolites. WG3 will reveal how beneficial microorganisms and pathogens interact with the metal metabolism of plants. Such interactions may, for example, involve changes in the uptake and intra-plant distribution of metals, enhanced or diminished expression or activity of metalloproteins.<br>Leader: Dr. Manuel Gonzalez-Guerrero <manuel.gonzalez@upm.es>,<br>deputy leader: Dr. Marek Vaculik <marek.vaculik@uniba.sk> |
| 10:00-10:30 | | ***coffee break*** |

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|---|---|---|
| 10:30-11:00 | **ALL members and supporters interested in this WG**, others can view posters / meet individually | **WG4 – Agronomy: Activity planning for GP2**<br>*MoU description:* Agronomic aspects of TM homeostasis. WG4 aims to conduct studies to contribute to better nutritional quality of food crops by using targeted fertilisation and agricultural management also in consideration of the new EU regulation on fertilisers, and to characterise crop plant response to low and excess application of TMs by employing physiological assays and analytical tools, which will be developed in WG1-3. WG4 will evaluate how micronutrient fertilisers are absorbed (after foliar vs. soil application, added in organic or mineral forms), how microorganisms and fungi can be used to improve plant TM nutrition (through a better use of soil metals), how TMs are transported within crop plants and how their localisation and speciation changes in this process. Finally, workshops / symposia will be organised for farmers associations and industrial stakeholders dealing with targeted breeding, fertilisation and micronutrient foliar fertiliser production. This will be done in the last year of the Action, to facilitate the transfer of the knowledge gained from the COST Action to the stakeholders.<br>Leader: Prof. Philip White <Philip.White@hutton.ac.uk>,<br>deputy leader: Dr. Valerie Bert <Valerie.BERT@ineris.fr> |
| 11:00-11:30 | **ALL members and supporters interested in this WG**, others can view posters / meet individually | **WG5 – Dissemination: Activity planning for GP2**<br>*MoU description:* Dissemination. WG5 will be focused on how to best target the results from this Action to the different stakeholders. WG5 will be responsible for preparing the press releases to news outlets, update the Action's social media accounts, and support the efforts at the national front. Moreover, WG5 will organise training sessions on writing skills to target research journals, farmer bulletins, and press releases. In addition, WG5 will coordinate any joint project by groups within the consortium to maximise group interactions, avoid overlaps and redundancy of research efforts, and increase success rates.<br>Leader: Prof. Ute Kraemer <Ute.Kraemer@ruhr-uni-bochum.de>,<br>deputy leader: Prof. Alexander Lux <alexander.lux@uniba.sk> |
| 11:30-12:00 | **ALL members and supporters interested in this WG**, others can view posters / meet individually | **WG6 – IPP: Activity planning for GP2**<br>*MoU description:* Intellectual property protection. It is expected that a number of the results obtained within the PLANTMETALS Action will have an impact on the private sector. WG6 will coordinate all the IP protection efforts, promoting them whenever possible in collaboration with the universities involved – (which usually have their own IP department). In addition, to maximise this output, WG6 will organise a training session on IP protection for the younger participants and promote meetings with the private sector to ensure the exploitation of the key results of the Action.<br>Leader: Dr. Martin Trtilek <martin@psi.cz>,<br>deputy leader: Prof. Mark Aarts <mark.aarts@wur.nl> |

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| 12:00-12_30 | | **Concluding remarks, farewell** |
| 12:30-14:30 | **ALL members and supporters** | ***lunch break – lunch served in canteen 13:00-13:30***<br>*afterwards: poster de-mounting, departure or self-organised activities* |
| 14:30-16:00 | **interested members and supporters** | **plant phenotypic workshop offered by Photon Systems Instruments** |
| | | |

## Alphabetial list of contributions - posters and talks

Every participant (also those giving a talk!) needs to present a poster. The posters need to:

1) be A0 in size,

2) be designed for portrait orientation,

3) have the logo of our COST Action (which you can download in our intranet!) in their title area,

4) have our COST Action mentioned in the "acknowledgements"

In addition, we encourage to have a photo of yourself on the poster as many of us don't know each other yet, and the photo will make finding an owner of a poster easier in our interactive poster session where naturally people will not stay all the time at their posters. However, by EU general data protection rules (GDPR), having a photo on your poster is completely VOLUNTARY!

List of all PLANTMETALS members incl. their WG subscriptions:

https://plantmetals.eu/plantmetals-members.html

| Presenting author | Organization | Title |
|---|---|---|
| Elisa Andresen | Biology Centre, Czech Academy of Sciences, Institute of Plant Molecular Biology, Department of Plant Biophysics & Biochemisty, Czech Republic | Chronic exposure of soybean plants to nanomolar cadmium reveals specific additional high-affinity targets of Cd toxicity |
| Emre Aksoy | Nigde Omer Halisdemir University, Turkey | Potential new regulators of Fe uptake in the roots: the GATA family |
| Abdel Rahman Al Tawaha | Al Hussein Bin Talal University, Jordan | Effects of foliar application of boron (B) on growth, yield and yield components and mineral composition of fenugreek |
| Marisa Almeida | CIIMAR, Portugal | Phytoremediation of contaminated soils aiming metal recovery |
| Esmira Alirzayeva | Azerbaijan National Academy of Sciences, Azerbaijan | Heavy metal tolerance mechanism of Artemisia plants |
| Karolina Barcauskaite | LAMMC, Lithuania | Fertilization Effect on Cannabis sativa L. mineral composition |
| Syed Nadeem Hussain Bokhari | Biology Centre, Czech Academy of Sciences, Institute of Plant Molecular Biology, Department of Plant Biophysics & Biochemisty, Czech Republic | Ultra-trace Metal Speciation Analysis by Coupling of Sector-Field ICP-MS to High-Resolution Size Exclusion and Reversed-Phase Liquid Chromatograph |
| Hester Blommaert | Université Grenoble Alpes, France | The Odyssey of Cd in Cacao Trees: from Soil to Bean |
| Jagna Chmielowska-Bąk | Adam Mickiewicz University, Poznań, Poland | ROS dependent events in soybean seedlings subjected to short term cadmium stress. |
| Valerie Bert *(participation not yet confirmed)* | INERIS, France | Agronomic Aspects of Trace Metal Homeostasis |
| Antonio Colussi | Biology Centre, Czech Academy of Sciences, Institute of Plant Molecular Biology, Department of Plant Biophysics & Biochemisty, Czech Republic | The role of metals in photosynthesis regulation of Trichodesmium. |
| Catherine Curie | BPMP Montpellier, France | Manganese transport and its regulation in Arabidopsis |
| Robert Dulfer | Biology Centre, Czech Academy of Sciences, Institute of Plant Molecular Biology, Department of Plant Biophysics & Biochemisty, Czech Republic | Cost Action Virtual Networking Grants as additional support during pandemic restrictions |
| Seckin Eroglu | Middle East Technical University, Turkey | Is iron involved in seed germination? |

| Presenting author | Organization | Title |
|---|---|---|
| Gerald Falkenberg | Deutsches Elektronen-Synchrotron DESY, Germany | Synchrotron Radiation X-ray Fluorescence Imaging Capabilities at Beamline P06 at DESY/Hamburg |
| Isvett Josefina Flores-Sanchez | Biology Centre, Czech Academy of Sciences, Institute of Plant Molecular Biology, Department of Plant Biophysics & Biochemisty, Czech Republic | Metalloproteomic Analysis from Soybean Roots Treated with Nanomolar Concentrations of Cadmium |
| Eva Freisinger | University of Zurich, Department of Chemistry, Switzerland | WG2 - Metalloproteins and -enzymes |
| Antonella Furini | University of Verona, Italy | Comparative analysis identifies micro-RNA associated with zinc homeostasis in Arabidopsis thaliana and metal hyperaccumulator Arabidopsis halleri |
| Tsanko Gechev | Center of Plant Systems Biology and Biotechnology, Bulgaria | Research in the Center of Plant Systems Biology and Biotechnology |
| Manuel Gonzalez-Guerrero | Universidad Politécnica de Madrid, Spain | MtFPN2 delivers iron for symbiotic nitrogen fixation. |
| Libor Grubhoffer | Biology Centre, Czech Academy of Sciences, Czech Republic | Welcome |
| Emmanuel Guillon | ICMR UMR CNRS 7312 - Reims Champagne Ardenne University, France | Environmental chemistry dedicated to the metal trace element fate |
| Marie-Theres Hauser | Department of Applied Genetics and Cell Biology, BOKU - University of Natural Resources and Life Sciences Vienna, Austria | Identification and characterization of a novel cell wall associated metal-binding peptide |
| Stefanie Höller | Martin-Luther-University Halle-Wittenberg, Germany | Overexpression of METAL TOLERANCE PROTEIN8 reveals new aspects of metal transport in Arabidopsis thaliana seeds |
| Muhammad Imran | Nouryon, Netherlands | Chelated micronutrient usage for overcoming crop deficiencies |
| Marie-Pierre Isaure | Université de Pau et des Pays de l'Adour, France | Use of combined synchrotron radiation techniques to study metal localization and speciation in plants |
| Jan Jansa | Institute of Microbiology, Academy of Sciences of the Czech Republic | Arbuscular mycorrhizal fungi as selective filters for nutrient flows into plants |
| Noelia Jaime-Pérez | Biology Centre, Czech Academy of Sciences, Institute of Plant Molecular Biology, Department of Plant Biophysics & Biochemisty, Czech Republic | Zinc-containing bacteriochlorophyll a ([Zn]-BChl a) protects the photosynthetic process of Acidiphilium rubrum from copper toxicity damage |
| Biljana Jordanoska Shishkoska | University “ St. Kliment Ohridski” – Bitola, "Scientific tobacco Institute ”- Prilep , Macedonia | The influence of soil properties on the accumulation and availability of trace elements into vegetative parts of the oriental tobacco |
| Weronika Jozwiak | PPC ADOB, Poland | ADOB. The Power of Science. |
| Jana Kohanová | Comenius University in Bratislava, Faculty of Natural Sciences, Department of Plant Physiology, Slovakia | Remediation of aquatic environment by dried algae |
| Anna Kokavcová | Comenius University in Bratislava, Slovakia | Copper and zinc accumulation, distribution, and tolerance in the roots of Pistia stratiotes (L.) and its potential for phytoremediation |
| Biljana Kukavica | Faculty of Natural science and mathematics, Mladena Stojanovića, Bosnia & Herzegovina | Secondary metabolites as Fe and Cu chelators in selected medicinal plant species |
| Hendrik Küpper | Biology Centre, Czech Academy of Sciences, Institute of Plant Molecular Biology, Department of Plant Biophysics & Biochemisty, Czech Republic | Introduction to the COST Action “Trace metal metabolism in plants – PLANTMETALS” and the scientific contribution of the workgroup of its chair |
| Andjela Kuvelja | Biology Centre, Czech Academy of Sciences, Institute of Plant Molecular Biology, Department of Plant Biophysics & Biochemisty, Czech Republic | Photosynthetic activity and element distribution in Capsicum annum leaves infected with Botrytis cinerea under different Mn and Zn supply |

| Presenting author | Organization | Title |
|---|---|---|
| Alexander Lux | Comenius University in Bratislava, Faculty of Natural Sciences, Department of Plant Physiology, Slovakia | Checkpoints of transport processes in plants facing toxic metals/metalloids and checkpoints of COST Action "PLANTMETALS" - CA19116 dissemination. |
| Irena Macek | University of Ljubljana, Biotechnical Faculty, Slovenia | Long-term experiments in research of arbuscular mycorrhizal fungal community ecology |
| Viktorija Maksimova | Goce Delcev University, Faculty of Medical Sciences, North Macedonia | An introduction and perspective interests into PlantMetals COST Action |
| Małgorzata Czernicka | University of Agriculturae in Krakow, Poland | Iodine priming for enhancement of tolerance to waterlogging stress in Cucumis sativus |
| Nadica Maltar-Strmečki | Ruđer Bošković Institute, Croatia | EPR spectroscopy as a Tool to Investigate PLANTMETALS |
| Stéphane Mari | INRAE, France | Iron speciation in seeds of Arabidopsis thaliana and dynamics of vacuolar remobilization during germination |
| Bastian Meier | Martin-Luther-Universität Halle-Wittenberg, Germany | Calcium signalling in barley |
| Michel Mench | INRAE (UMR BIOGECO INRAE 1202), France | Foliar Si and Se supply, alone and combined with organic soil amendments, to promote crop production on metal(loid)-contaminated soils |
| Sylvain Merlot | CNRS Université Paris Saclay, France | Wide cross-species RNA-Seq comparison reveals convergent molecular mechanisms involved in nickel hyperaccumulation across dicotyledons |
| Adrien Mestrot | University of Bern, Switzerland | Speciation of trace elements in soils and plants |
| Tijana Milićević | Institute of Physics, Environmental Physics Laboratory, Serbia | Bioavailability, environmental and human health risk of potentially toxic elements in the soil-grapevine system |
| Ana Mijovilovich | Biology Centre, Czech Academy of Sciences, Institute of Plant Molecular Biology, Department of Plant Biophysics & Biochemisty, Czech Republic | What X-rays can tell about metals in plants: localization and chemistry. Current and future work. |
| Tomica Mišljenović | University of Belgrade, Faculty of Biology, Serbia | Bioaccumulation of potentially toxic elements in Noccaea kovatsii and N. praecox (Brassicaceae) from different geological substrates and their physiological responses to Ni |
| Filis Morina | Biology Centre, Czech Academy of Sciences, Institute of Plant Molecular Biology, Department of Plant Biophysics & Biochemisty, Czech Republic | Beneficial role of zinc against biotic stress in metal non-hyperaccumulating plants |
| Levent Ozturk | Sabanci University, Tukrey | Interactive effects of cadmium and salt treatments on cadmium uptake of the heavy metal hyperaccumulator Thlaspi caerulescens |
| Veselin Petrov | Center of Plant Systems Biology and Biotechnology, Bulgaria | The PlantaSYST project: establishment of a new Center-of-Excellence in Bulgaria for the translation of fundamental research into sustainable bio-based technologies |
| Dorina Podar | Babes-Bolyai University, Romania | Plant metal transporters and plant-microorganisms interactions within the rhizosphere for heavy metal remediation and biofortification purposes |
| Giancarlo Renella | University of Padua, Italy | Contribution of the UniPd partner to the activities of the PLANTMETAL COST Action |

| Presenting author | Organization | Title |
|---|---|---|
| Siniša Škondrić | Faculty of Natural Sciences and Mathematics<br>Banja Luka, Bosnia & Herzegovina | Secondary metabolites as Fe and Cu chelators in selected medicinal plant species |
| Petr Soudek | Ústav experimentální botaniky AV ČR, Czech Republic | Toxicity versus essentiality – two faces of transient elements in plant metabolism |
| Sylwester Smoleń | Faculty of Biotechnology and Horticulture, University of Agriculture in Krakow, Poland | Anticancer potential of lettuce biofortified with iodine in human gastrointestinal cancer cell lines as well as aspects of plant-derived thyroid hormone analogs in lettuce |
| Geraldine Sarret | Université Grenoble Alpes, France | Processes controlling metals in soil-plant systems: Interest of combining speciation and stable isotopes fractionation |
| Josef Špak | Biology Centre, Czech Academy of Sciences, Institute of Plant Molecular Biology, Czech Republic | <no presentation, director of Institute of Plant Molecular Biology> |
| Tatiana Stratulat | Institute of Genetics, Physiology and Plant Protection, Moldova | Quantification of metals content in vegetables and fruits cultivated in Moldova and health risk assessment |
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| Liliana Tudoreanu | University of Agronomic Sciences and Veterinary Medicine Bucharest Bucuresti, Romania | The effect of soil cadmium concentration on the nutritional value of Brasiliana Lettuce plants |
| Luiza Usevičiūtė | Vilnius Gediminas Technical University, Lithuania | Effect of pine wood biochar on immobilisation of potentially toxic elements in contaminated soil and uptake by oat (Avena Sativa L.) |
| Marek Vaculik | Plant Science and Biodiversity Centre, SAS, Slovakia | Alleviation of metal(loid) toxicity in plants by silicon |
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| Verbruggen | Université libre de Bruxelles, Belgium | Different strategies of Cd resistance in Arabidopsis halleri |
| Milada Vítová | Institute of Microbiology of the Czech Academy of Sciences, Czech Republic | Red mud as a secondary source of scarce metals - Recovery using red microalgae |
| Matthias Wiggenhauser | ETH Zurich, Switzerland | 'Let's label! The use of enriched metal stable isotopes in soil-fertilizer-crop systems' |
| Muhamet Zogaj | University of Prishtina, Faculty of Agriculture and Veterinary, Kosovo | Heavy metal contents in agricultural soils of Kosovo |

## Alphabetial list of Abstracts -

- This list contains all talks, as well as those poster contributions that voluntarily supplied an abstract

– The presenting author is underlined

- Texts and affiliations are as supplied by the authors, no corrections applied

- Countries were added to the affiliations according to the e-COST profiles of the participants

- The e-mail of the presenting author was added according to the e-COST profiles of the participants

# Chronic exposure of soybean plants to nanomolar cadmium reveals specific additional high-affinity targets of Cd toxicity

Elisa Andresen (1), Lyudmila Lyubenova (1), Tomáš Hubáček (2), Syed Nadeem Hussain Bokhari(1), Šárka Matoušková (3), Ana Mijovilovich (1), Jan Rohovec (3) and Hendrik Küpper (1,4)

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Solving the global environmental and agricultural problem of chronic low-level Cd exposure requires better mechanistic understanding. Here, soybean (Glycine max) plants were exposed to Cd concentrations ranging from 0.5 nM (background concentration, control) to 3 µM. Plants were cultivated hydroponically under non-nodulating conditions for 10 weeks. Toxicity symptoms, net photosynthetic oxygen production, photosynthesis biophysics (Chl fluorescence: Kautsky and OJIP), Cd binding to proteins (metalloproteomics by HPLC-ICPMS) and Cd ligands in LHCII (XANES), accumulation of elements, Chl and metabolites were monitored.
There was a distinct threshold concentration of toxicity onset (140 nM) as visible from the strongly decreased growth, the switch-like pattern for nutrient uptake and metal accumulation and photosynthetic fluorescence parameters like ΦRE10 (OJIP) and saturation of the PS. XANES analyses of isolated LHCII revealed that Cd was bound to N or O (and not S) atoms. Nutrient deficiencies due to inhibited uptake could be caused by transporter blockage by Cd ions. The changes in specific fluorescence kinetic parameters indicate electrons not being transferred from PSII to PSI. Inhibition of photosynthesis combined with inhibition of root function could explain why the amino acid and carbohydrate metabolism decreased in favour of molecules involved in Cd-stress tolerance (e.g. anti-oxidative system, detoxifying ligands).

Acknowledgement: This work was supported by COST (CA 19116 “Trace metal metabolism in plants – PLANTMETALS), the Ministry of Education of the Czech Republic with co-financing from the European Union (grant KOROLID, CZ.02.1.01/0.0/0.0/15_003/0000336), and the Czech Academy of Sciences (RVO: 60077344).

Potential new regulators of Fe uptake in the roots: the GATA family

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Iron (Fe) is one of the essential micronutrients for plants, and Fe deficiency is among the most widespread nutritional deficiencies. Although mechanisms involved in Fe uptake and root-to-shoot translocation are well-known, transcription factors controlling these mechanisms are limited to some families. Our bioinformatics studies suggested the involvement of two members of the GATA family, namely GATA9 andGATA12, in the regulation of Fe signaling in plants. GATA transcription factors are controlled by different hormones and thus play important roles in plant development and stress responses. Therefore, we investigated the potential functions of these GATA proteins in Fe uptake and distribution in Arabidopsis thaliana. gata9-1, gata12-1, and gata12-2 mutants showed less chlorosis than the Col-0 while the chlorosis level was much higher inGATA12 overexpression (OE) lines under Fe deficiency. The increase in ferric chelate reductase (FCR) activity was higher in gata9-1 and gata12-1 mutants as compared to Col-0 under Fe deficiency whereas it was similar in gata12-2. Interestingly, the FCR activity was increased much less in GATA OE lines than it was in Col-0. Fe-uptake genes were significantly upregulated in GATA OE lines, except for AHA2. The phenotype of single mutants was significantly enhanced in the gata9-1gata12-1 mutant under Fe deficiency. Taken together, these data suggest the essential roles of GATA family members in Fe uptake in plant roots.

Acknowledgements: This research is supported by The Scientific and Technological Research Council of Turkey (project number 119Z149) and the European Union (COST project number 19116).

# Effects of foliar application of boron (B) on growth, yield and yield components and mineral composition of fenugreek

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"Fenugreek seeds are high in minerals, vitamins, and flavonoids, and they play an important role in human health. When boron (B) demands exceed what the soil can supply, foliar application may be used to supply B to a crop. A study was carried out to determine the response of fenugreek to boron fertilizer. Fenugreek grown plots were sprayed with boron (0, 50, 100 and 150 mg L−1) in a randomized complete block design. The measured variables were seed yield plant-1, number of pods plant-1, number of seeds pod-1, plant height (g), pod length, number of branches per plant, number of leaves per plant. Results testified that fenugreek responded well to foliar application to (B) in term of yield and related traits along with mineral accumulation. The control treatment (no B application) had the lowest grain yield which was significantly lower than the yield observed with the foliar application of (B) in various treatment. In conclusion fenugreek yield and yield components was affected due to grain set failure induced by boron deficiency and it was possible to overcome this element deficiency by soil application at 150 mg L−1

Agronomic Aspects of Trace Metal Homeostasis

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Appropriate crop nutrition addresses many of WHO's Sustainable Development Goals by reducing hunger, promoting health, conserving resources and reducing environmental damage. Some trace metals (TMs), such as Fe, Zn, Mn, Cu, Ni and Mo, are nutrients required by both plants and animals, although their excess can be toxic. Other TMs, such as Cd, Pb and Hg, are simply hazardous to health. Working Group 4 will investigate agronomic aspects of TMs in cropping systems. The aim is to develop strategies to improve both yields and nutritional quality of edible crops whilst minimising environmental impacts (T4.1). Studies will characterise crop responses to TMs (T4.2), the uptake of TMs from soil or foliar fertilisers (T4.3), the influence of microbes on plant acquisition of TMs (T4.4), and the movement and accumulation of TMs within the plant (T4.5). This talk will provide examples of such studies.

# The Odyssey of Cd in Cacao Trees: from Soil to Bean

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The accumulation of the toxic metal Cd in cacao beans has recently become a subject of intense research after the European Union lowered its limits in chocolate. Genetics-based techniques may offer sustainable strategies for Cd mitigation in cacao. Nonetheless, this reduction potential is yet to be exploited since there is a lack of knowledge on how Cd is transported from soil to the cacao bean. Here, we aimed to compare the mechanisms that control Cd transfer in two genotypes by a combination of Cd stable isotope analyses and X-ray absorption spectroscopy. Three replicate trees of a low (L) and a high Cd accumulating (H) cultivar were selected from a conservatory of cacao cultivars in Trinidad (soil Cd: 0.3 mg kg-1). Cadmium concentrations in leaves and beans of H were factor 3 higher than in L. Both cultivars showed a similar partitioning strategy, i.e. Cd concentrations increased in the order: placenta < nib < testa < pod husk < root < young leaf - old leaf < wood. In roots, leaves, and wood tissues of H, Cd was mainly bound to carboxyl ligands, while in the nib Cd was mostly associated with phytate. The first isotope measurements in H indicate a fractionation between organs. Beans were enriched in light isotopes compared to the leaves. This suggests different pathways and types of ligands for Cd in cacao compared to cereal grains. More isotope data on other tissues are currently measured, and will further unravel the pathways of Cd in the two genotypes.

Ultra-trace Metal Speciation Analysis by Coupling of Sector-Field ICP-MS to High-Resolution Size Exclusion and Reversed-Phase Liquid Chromatograph

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Investigation of trace metals bound to macromolecules and pigments require methods of metal speciation analysis in complex matrices and online quantification of matrix elements with sub nanomolar detection limits. We present a method development and optimization of such metal analysis with a custom built HPLC-ICP-MS system that mainly included metal-free HPLC-DAD system and sector-field ICP-MS detection (ICF-sfMS) with desolvating injection and optimization of sample handling. Protein were identified with SEC (size exclusion chromatography) and AEC (anion exchange chromatography) using ammonium bicarbonate buffer. Analysis of metal exchange in pigments was performed by reversed-phase chromatography with a methanol-acetone gradient via organic matrix interface (RPC-ICP-sfMS). The optimized method has detection limits in the picomolar range in protein buffer only limited by purity of chemicals. Examples for Cd binding to soybean proteins and chlorophyll, Cr binding to Arabidopsis thaliana proteins, La binding to Desmodesmus quadricauda proteins, and Cu binding to Rhodospirillum rubrum proteins and pigments demonstrate the sensitivity at background concentration levels of solutions made from analytical grade chemicals. The tested method is being applied in different metal speciation analysis projects.

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ROS dependent events in soybean seedlings subjected to short term cadmium stress.

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Reactive oxygen species (ROS) are recognized as crucial elements in plants response to metals. In access these molecules lead to oxidative stress and damage of various cellular elements. On the other hand certain ROS level is required for signalling events and stress response.
The aim of present study is elucidation of the role of ROS in the early response of soybean seedlings to Cd, with particular emphasis on oxidative modifications of transcripts. The soybean seedlings were exposed to Cd at the concentration 10 mg/l for 1, 2 and 3 h. Thereafter, the level of superoxide anion, hydrogen peroxide, lipid peroxidation, protein carbonylation and most common RNA oxidative modification, 8-hydroxyguanosine (8-OHG), have been assessed.
The results indicate that 8-OHG formation is one of the earliest ROS-dependent events in the response of soybean seedlings to Cd. The results also show that oxidation of transcripts is at least partially mediated by ROS derived from mitochondria. Other studies carried out on animal and plant models evidenced that RNA oxidation is a selective process and that it can be engaged in post-transcriptional regulation of genes expression.
Future research carried out in frame of present project will include intracellular localization of oxidized transcripts and their identification by the means of next-generation sequencing.

Acknowledgement: The research was financed by National Science Center, Poland, in the frame of the project number 2019/33/B/NZ9/00058.

Mn transport and its regulation in Arabidopsis

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Mn deficiency is a widespread nutritional disorder that causes substantial yield losses and decreases the quality of edible plant parts. Little is known regarding the molecular mechanisms that control Mn homeostasis. We have shown that Arabidopsis growth in Mn limiting conditions relies on a high affinity Mn uptake transport system encoded by NRAMP1 (Cailliatte et al. 2010 Plant Cell 22 : 904-917). We have also reported that IRT1, the iron uptake transporter at the root surface, cooperates with NRAMP1 for Mn acquisition (Castaings et al. 2016 Sci. Rep. 6, 37222). Unlike numerous high-affinity transporters, NRAMP1 expression is only little regulated at the transcriptional level. We have recently discovered that NRAMP1 responds to Mn availability by trafficking dynamically between the plasma membrane and various endomembrane compartments. In particular, we show that NRAMP1 endocytosis from the plasma membrane is controlled by phosphorylation of the protein. A mutagenesis strategy has enabled us to link phosphorylation of specific amino acid residues with membrane localization of NRAMP1 and with tolerance of the plant to Mn exposure (Castaings et al. 2021 Plant J. doi: 10.1111/tpj.15239). We are currently attempting to identify NRAMP1 protein partners through MS and genetic approaches.

Is iron involved in seed germination?

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Iron (Fe) joins in distinct physiological processes such as photosynthesis, flowering time, and programmed cell death in plants. However, whether it is involved in germination is not known. Recently, Murgia and Morandini showed seeds from Fe deficient plants had stronger dormancy, an inner property of seed that prevents germination despite how optimum the conditions are. Furthermore, Fe is a significant source for reactive oxygen species (ROS), while ROS is central to seed germination. ROS levels signal when to break dormancy, involved in loosing endosperm -called endosperm weakening-, and radicle protrusion.
We have observed a late germination phenotype in a mutant Arabidopsis thaliana line that cannot synthase one of the major chelators for Fe in plants. The phenotype could be rescued by exogeneous application of the chelator and also Fe supplementation, suggesting a possible link between Fe and germination.

Acknowledgement: I acknowledge TUBITAK (118Z788) for the financial support for this research.

# Metalloproteomic Analysis from Soybean Roots Treated with Nanomolar Concentrations of Cadmium

Isvett Josefina Flores-Sanchez (1), Elisa Andresen (1), Syed Nadeem Hussain Bokhari (1), Hendrik Küpper (1,2)

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Cadmium (Cd) is an environmental pollutant in agricultural soils since the uptake of Cd by plants represents an entry point into the food chain. Roots are the main organ for the uptake of nutrients and toxic metals. Previous studies have shown the presence of Cd-binding proteins in leaves from soybean (Glycine max, var. Erin) plants exposed to nanomolar concentrations of Cd. Metalloproteomics is an emerging technique for identifying the binding of metals and metalloids to proteins within a cell or tissue type. Inductively coupled plasma mass spectrometry (ICP-MS)-based metalloproteomics is an analytical platform which has been used for identification of putative metal/metalloid proteins within biological systems. In this study, soybean (G. max, var. Galina) plants were fed using a continuous-flow solution culture with 20 nM and 1000 nM Cd. Purification of Cd-binding proteins from roots was performed by 3 chromatographic methods (anion exchange chromatography with 2 different columns and size exclusion chromatography) coupled to ICP-MS. Our preliminary results shown the identification by liquid chromatography-tandem mass spectrometry (LC-ESI-MS/MS) of the membrane metalloenzymes allantoinase (EC 3.5.2.5) and peroxidase (EC 1.11.1), as well as the transmembrane proteins aquaporins. Further analyses are carried out in order to identify soluble Cd-binding proteins from soybean roots.

Acknowledgement: This work was supported by COST (CA 19116 “Trace metal metabolism in plants – PLANTMETALS), the Ministry of Education of the Czech Republic with co-financing from the European Union (grant KOROLID, CZ.02.1.01/0.0/0.0/15_003/0000336), and the Czech Academy of Sciences (RVO: 60077344).

WG2 - Metalloproteins and -enzymes

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"WG2 is committed to the identification and study of metalloproteins important for trace metal homeostasis, hence the uptake, transport, cellular distribution, and speciation of these metals. Further, a focus is set on the interaction of trace metal binding proteins and enzymes with biological ligands and chelators as well as how the specificity towards the respective target metal ion is achieved. Other task of WG2 to be specifically highlighted are the determination of affinities and (3D) structural details of metal ion coordination as well as turn-over rates of metal-dependent enzymes.
This contribution will introduce the current members of WG2, their research focus, and skills as well as instrumental expertise they can provide for the Action. In addition, collaborations that have been already initiated will be presented and current research needs of members phrased to help start new and vivid interactions to help fulfilling the goals of the Action."

# Comparative analysis identifies micro-RNA associated with zinc homeostasis in Arabidopsis thaliana and metal hyperaccumulator Arabidopsis halleri

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miRNAs are key players in mineral homeostasis, both in the control of nutrient balance and in the response to toxic trace elements. However, the effect of Zn excess on miRNAs has not been elucidated; moreover, no data are present regarding miRNAs in hyperaccumulator species, where metal homeostasis is tightly regulated. In this work expression levels of mature miRNAs were measured by RNA-Seq in Zn-sensitive Arabidopsis thaliana grown in control onditions and upon high Zn in soil and in Zn hyperaccumulator Arabidopsis halleri in control conditions. Differential expression of notable miRNAs and their targets was confirmed by real-time RT-PCR. The comparison in A. thaliana revealed a small subset modulated upon Zn treatment, that is associated with stress response and nutrient homeostasis. On the other hand, a more consistent group of miRNAs was differentially expressed in A. halleri in comparison with A. thaliana, reflecting inherent differences in nutritional requirements and response to stresses, as well as in plant growth and development. Overall, these results confirm the involvement of miRNAs in Zn homeostasis and support the hypothesis of distinct regulatory pathways in hyperaccumulator species.

# ADOB. The Power of Science.

Adam Nawrocki, Weronika Jóźwiak

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ADOB is a well-established fertiliser manufacturer based in Poland. We have been producing innovative fertilisers, nourishing plants and enhancing the productivity of agricultural and horticultural crops for almost 30 years, in Poland and in over 80 other countries across five continents. Based on unique and patented technologies our fertilisers are developed in cooperation with leading commercial and research institutions and produced with state-of-the-art, hi-tech expertise. Our product portfolio includes single, oligo and multi-nutrient fertilisers in liquid, crystalline and microgranular formulations. They are specifically designed for both indoor and outdoor application and can be used on field, orchard, vegetable, energy, ornamental, herb and nutraceutical crops. The fertilisers can be applied using all current application methods, including foliar spray, fertigation, hydroponics, drenching, seed-treatment, seed-priming, and base and top/side soil dressing. Our innovative "2.0" technology significantly improves nutrient uptake and availability, enhancing crop performance. We continually improve our production processes, thereby filing and acquiring technological patents. We own modern, excellently equipped laboratories and an R&D centre. Our team of over 20 highly qualified specialists are continuously developing new products and optimising the effectiveness of fertilisers in order to enhance the genetic potential of important crop plants.

WP3: Responses of plant Trace Metals to the environment

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The environment greatly affects plant growth and processes related to their development. One of the current problems of agriculture is the depletion important trace metals in soils, while other sites suffer from to high input of toxic trace metals, especially due to high industrialization or wrong soil management. Therefore, in WP3 we will focus on how plant metabolism is modulated to ensure sufficient metal uptake when growing in deficient soils or detoxify trace metals when present at excess levels. Moreover, this WP will address the challenge of climate change by determining how drought, high CO2, and other abiotic stresses affect plant metal homeostasis. Since plants are in a tight contact and relationship with other environmental biotic factors, the role of plant-associated microorganisms will also be considered. Beneficial microorganisms and plant pathogens will be in a focus of WP3 either as means to improve metal uptake and ameliorate metal toxicity, as metal sinks for environmental-friendly solutions for sustainable agriculture, or as diseases to combat where metals play a role in plant immunity. These are tasks in which synergies with the other WPs will be established to succeed.

MtFPN2 delivers iron for symbiotic nitrogen fixation.

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Legumes overcome nitrogen limitation by establishing an endosymbiotic relationship with diazotrophic bacteria. Within organelle-like structures in root nodule cells, the symbiosomes, these bacteria synthesize the enzyme nitrogenase that converts N2 into NH3 to be transferred to the host plant. For symbiotic nitrogen fixation to work, large amounts of iron have to be delivered to the symbiosomes. Here we present Medicago truncatula FPN2, a nodule-specific iron efflux protein. Promoter:gus fusions and protein immunolocalization studies showed that MtFPN2 was located in an endomembrane compartment in nodule vascular cells and in the symbiosomes of cells in the differentiation to early fixation zones of the nodule. Loss of MtFPN2 function resulted in a reduction of biomass production when the plant obtained its nitrogen exclusively from symbiotic nitrogen fixation. This was caused by the loss of nitrogenase activity in nodules, the consequence of iron not reaching this compartment. X-ray fluorescence analyses and XANES studies showed that indeed iron distribution and speciation in this organ was altered by MtFPN2 loss. This phenotype was restored when MtFPN2 expression was driven by a promoter specific of the differentiation-fixation zones, while no improvement was observed when a vascular promoter was used. These data are consistent with a model in which MtFPN2 transports iron into symbiosomes of nitrogen-fixing cells.

Acknowledgement: Work supported by ERC Starting Grant ERC-2013-StG-335284

Identification and characterization of a novel cell wall associated metal-binding peptide

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Trace metals (TMs) are both ubiquitous and crucial in biology, but, if present in excess, are sources of substantial environmental and health problems. On the other hand, organisms such as hyper-accumulating plants even flourish in environments containing high levels of TMs due to their efficient detoxification system typically involving several TM-binding and transport molecules including peptides. By analysing Salix caprea clones from metallicolous and non-metallicolous areas with different TM accumulation characteristics genes were isolated that were upregulated upon long term exposure of elevated Zn and/or Cd concentrations. Remarkably, several of these genes were putatively secreted and/or associated to cell wall functions. We present the molecular characterisation of one of these genes coding for a novel TM-binding peptide from S. caprea. TM-binding was shown with the purified recombinantly expressed peptide and size exclusion (SEC)-ICP-MS. Cross-species experiments in the model plant Arabidopsis thaliana revealed that the processed peptide is secreted into the cell wall. Receptors of closely related peptides of A. thaliana are involved in cell wall integrity signalling and growth responses upon elevated levels of TMs. We will present a model how these peptides might act together to mediate growth adaptation to TMs.

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# Overexpression of METAL TOLERANCE PROTEIN8 reveals new aspects of metal transport in Arabidopsis thaliana seeds

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The distribution of metals in seeds is essential during seed development and germination, and its understanding is especially important in terms of biofortification.
In dry seeds of Arabidopsis thaliana, manganese (Mn) localization is defined to subepidermal cells at the abaxial side of the cotyledons, while iron (Fe) accumulates mainly around the vasculature. The vacuolar importer VIT1 is responsible for the Fe localization pattern, while the vacuolar Mn transporter METAL TOLERANCE PROTEIN 8 (MTP8) mediates Mn allocation to specific cell types in the developing embryo, and Fe re-allocation as well as Mn tolerance during imbibition.
In this study, we used synchrotron X-ray fluorescence (µ-XRF) to analyse if an overexpression of MTP8 driven by the CaMV 35S promoter has an effect on Mn tolerance during imbibition and on Mn and Fe storage in seeds. In contrast to the expected homogenous distribution, seeds overexpressing MTP8 (35S:MTP8) accumulated Mn and Fe at the same sites as in wild type seeds. However, overexpression of MTP8 enhanced Mn tolerance of germinating seeds during imbibition, pointing to an additional role of this transporter in Mn detoxification at this early developmental stage.
The results demonstrate that MTP8 expression pattern do not determine Mn or Fe localization in mature seeds primarily. MTP8 activity might be regulated post-translationally, or metals might be available only at cell-specific sites as determined by upstream transport processes.

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# Chelated micronutrient usage for overcoming crop deficiencies

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As the rising world population reduces the amount of arable land available for food production, the cultivation of less fertile soils has become essential. This has made the ability to deliver nutrients to crops in an efficient, environmentally safe way extremely important. To make the most of the land we have, we must use innovative methods to preserve it. Through advanced hydroponic and fertigation techniques using specialized micronutrient fertilizers, we can save huge amounts of water and address water scarcity issues in many agricultural areas.

Micronutrients such as iron, manganese and zinc are crucial to the health of plants and crops. They help improve crop quality and generate higher yields around the world.

Nouryon's chelated micronutrients are highly effective for a variety of applications, such as fertigation to soil, foliar, hydroponics, and seed coating.

Chelating agents keep metals in solution and therefore plant available. The chelating agent surrounds the metal ion — like iron, manganese or zinc — and prevents it from reacting with phosphates, carbonates or hydroxides which would cause precipitation of the metal. Plants are able to use the metals from the chelate. In this way, fewer metals need to be applied to avoid fixation and loss of micronutrients in the soil.

Arbuscular mycorrhizal fungi as selective filters for nutrient flows into plants

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Arbuscular mycorrhizal fungi establish an intimate association with roots of majority of extant plant species. They are nourished by reduced carbon compounds by their host plant and provide a multitude of benefits to its host in return. Besides efficient acquisition, long-distance transport, and supply to plants of orthophosphate from the soil solution it has recently been documented that the fungi do obtain a significant share of phosphorus and nitrogen from organic sources, in concert with activity of other soil microorganisms. They also are capable of efficient acquisition and supply to plants of micronutrients such as zinc and copper, whereas they block uptake via roots of excessive amounts of cadmium, cesium and zinc. Knowledge on molecular mechanisms of micronutrient uptake by the fungi and further exchange (or reduction thereof) with the plants still needs to be improved, particularly with respect to temporal dynamics and environmental context. Two widely neglected scenario are stressed here, namely the gradient of nutrient availabilities and the phenomenon of common mycorrhizal networks, when the same fungal colony interconnects and redistributes resources between two or more neighbouring plants at the same time. Depending on the conditions, the outcome of the symbiosis for any single plant could span gradient of mutualism to parasitism. This should carefully be considered when thinking about alternative agricultural models with limited or no inputs.

The influence of soil properties on the accumulation and availability of trace elements into vegetative parts of the oriental tobacco

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I have been employed as a senior research associate at Department of agrotechnics at Scientific tobacco institute-Prilep. Through the systematic work of the Institution overall we make systematic control of fertility of tobacco soils, we create new tobacco varieties and preserve and improve the ones already registered, optimize agricultural practices, investigate diseases, pests and weeds on tobacco, study the chemical composition of tobacco and tobacco products, its post-harvest processes and study the economic problems in tobacco industry. This Institution has close relations with tobacco enterprises and by concrete activities it takes active participation in solution of the problems in tobacco production which is especially important for the Macedonian economy. The scientific-research work in my department is based on study through field demonstrative experiments on targeted fertilization and optimization of agricultural practices, investigations of the major macro-and micro-elements in the soil and in the tobacco raw, important water-physical characteristics of the soil and sets regimes for tobacco irrigation. Main part of my research interest is to determine the intensity of accumulation, uptake of various elements by vegetative organs of oriental tobacco and to determine possible relationships between certain chemical and physical properties of soil (pH, clay, cation exchange capacity, organic matter, and total organic carbon).

# Remediation of aquatic environment by dried algae

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Decontamination of aquatic environment represents a serious problem already a long time. The situation becomes critical, the news about contaminations of both marine and fresh waters appear almost daily. The traditional methods of removal of pollutants using chemical precipitation, coagulation, ion exchange, liquid extraction and filters are costly and often inaccessible. Methods of bioremediation may represent alternative way how to solve these problems. Other possibility is the use of biosorbents, this alternative was performed in the present study. Dried algae Chlorella sp. (Chlorophyta) were applied to a native sample obtained by blast furnace slag mineralization. In addition, a model sample prepared from a certified standard was used for the chromium experiment. The metabolism-independent adsorption and absorption of metal ions by cell walls is usually a very rapid process, and relatively high concentrations of metals can be sorbed. The dried alga used in our study has been shown as a rapid biosorbent, in relatively short time intervals. In addition, the results show that in the process of decontamination there was not only the binding of chromium to the cell surface but also its penetration through the cell wall.

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# Copper and zinc accumulation, distribution, and tolerance in the roots of Pistia stratiotes (L.) and its potential for phytoremediation

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The aquatic macrophyte Pistia stratiotes (L.) is an invasive species, widespread mainly in tropical and subtropical climates. In recent years it became common in standing waters of Central Europe. It can effectively accumulate Cd, Cr, and Pb, thus having the phytoremediation potential for wastewater treatment. Pistia has a specific type of the root cap - a root pocket which differs from the root cap of terrestrial plants. Due to the adaptation to the aquatic environment, the root pocket may have a specific function in root apex protection. The aim of this study was to determine the effects of environmentally relevant concentrations of Cu (0.1, 0.3, and 1 μM) and Zn (0.3, 1, and 3 μM) on root growth, photosynthetic activity, and element distribution in the root cap and the root apex of P. stratiotes. Using micro-X ray fluorescence, we observed that the highest concentrations of Cu and Zn affected their distribution in the Pistia roots in comparison to the controls. Increasing Cu concentration decreased the distribution of Zn. In addition, the differences in distribution of Fe and Mn were observed, they were mainly attached to the cells of the root cap whereas Cu and Zn were attached to the root proper. Direct measurements of chlorophyll fluorescence kinetics (OJIP) showed photosynthesis inhibition only in the highest Cu and Zn treatments.

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# Secondary metabolites as Fe and Cu chelators in selected medicinal plant species

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The transition metals Fe and Cu, although biogenic, can lead to the formation of one of the most reactive free radical, the hydroxy radical (.OH), in the Fenton reaction. Free radicals can disrupt the structure of biomolecules and the functions of the cell and are in the basis of many diseases. Phenolic compounds, as the most important secondary metabolites of plants, have the ability to remove free radicals in direct reactions, enzymatically catalyzed reactions or by chelating Fe and Cu. The aim of this study was to examine the ability of plant phenolic compounds isolated from selected medicinal plants: Salvia officinalis L., Trifolium pratense L., Agrimonia eupatoria L, Cichorium intybus L. and Vinca minor L. from the area of the Republika Srpska to chelate Fe and Cu, also their antioxidant activity and anti-diabetic activity as the ability to inhibit α-amylase activity. The results showed that ethanolic extract of A. eupatoria has the highest content of phenols and flavonoids, the highest antioxidant activity compared to ABTS, the highest chelating capacity of Cu as well as the highest antidiabetic activity. The highest Fe chelating ability was measured for V. minor extract. The conclusion of the paper would be that the ability to chelating Cu is related to the total content of phenolic compounds and flavonoids while the ability to chelating Fe is the result of the presence of specific phenolic compounds that have high chelating ability. Additional research is also needed to find out whether there are clear correlations between antioxidant activity, transition metal chelating ability, and antidiabetic activity of plant extracts.

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# Introduction to the COST Action “Trace metal metabolism in plants – PLANTMETALS” and the scientific contribution of the workgroup of its chair

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Trace metals (TMs, e.g. Cu, Fe, Mn, Mo, Ni, Zn) are essential for organisms as active centres of enzymes, as about one third of all proteins are metalloproteins. Therefore, TM homeostasis in plants is at the core of many challenges that agriculture and human society are currently facing. As explained in detail in our memorandum of understanding (MoU) the COST Action PLANTMETALS tackles basic and applied issues related to trace metal deficiency or excess in plant physiology and crop production by the combined expertise of physiologists, (bio)physicists, (bio)(geo)chemists, molecular geneticists, ecologists, agronomists and soil scientists. Knowledge will be translated to the needs of farmers and consumers, with inputs from companies. The talk will illustrate how our Action wants to achieve this, and it will review the development and activity of the PLANTMETALS Action so far.
Besides the coordinating role, my group will contribute to this Action by fundamental research aiming to improve the mechanistic knowledge of metal deficiency/toxicity stress, the role of trace elements in plant defence response to pathogens, and metal metabolism in general. This is done in diverse photosynthetic organisms with emphasis on crops and metal hyperaccumulator model plants, mostly using biochemical and biophysical methods. This opens many possibilities for collaboration within this COST network, some of which are already ongoing. The talk will show some key examples of our research.

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Checkpoints of transport processes in plants facing toxic metals/metalloids and checkpoints of COST Action "PLANTMETALS" - CA19116 dissemination.

Along the plant body there are several checkpoints to control the transport processes. These checkpoints may have a crucial function in regulation of water transport, movement of various ions, including toxic metals, metalloids, or excessive concentration of even essential elements. The study of these checkpoints represents an important part of research activities of our team at the Comenius University in Bratislava. A summary of this research will be introduced.
For the dissemination of our COST Action "PLANTMETALS“, there are also several important checkpoints. Some of them can be marked as “fulfilled”. These are logo, twitter link (prepared by Ute), webpage with mailing list (prepared by Hendrik as chair of the Action). Very useful is also intranet (prepared by Grant manager Robert Dulfer and IT specialist Zdenek Peltan). The list of checkpoints “to be done” remains long, and we should take the opportunity of this meeting to jointly discuss these here. Among them is the production of a flier brochure that informs about our CA 19116 activities. There is a budget for open access publications and for STSMs, which should be spent soon for this period. We should consider several points of our objectives as written in the MoU.

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# Long-term experiments in research of arbuscular mycorrhizal fungal community ecology

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The impacts of long-term changes in abiotic factors on soil microbiota remain largely unknown. This is particularly true for the arbuscular mycorrhizal fungi (AMF), which form mutualistic associations with over 2/3 of terrestrial plants. We used NGS to quantify the response of AMF communities to long-term changes in environmental factors in two different systems: (1) a long-term free-air carbon dioxide enrichment (FACE) experiment (Giessen, DE); and (2) natural CO2 springs or mofettes with soil hypoxia induced by geogenic CO2. In the FACE eCO2 significantly increased AMF richness but had a less-pronounced impact on the composition of their communities. However, while broader changes in community composition were not observed, more subtle responses of specific AMF taxa were with populations both increasing and decreasing in abundance in response to eCO2. On the other hand, a complete AMF community turn-over was found in areas exposed to long-term geogenic CO2 in mofettes that was consistent through several years. This suggests that the temporal dynamics of AMF communities may be disturbed by anthropogenic stressors, however a distinct level of responses happens depending on the intensity of the stressor and its nature of action. As AMF are functionally differentiated, with different taxa providing different benefits to host plants, changes in communities in response to abiotic factors may significantly impact terrestrial plant communities, mineral nutrition and productivity.

# An introduction and perspective interests into PlantMetals COST Action

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Republic of North Macedonia is a small country located on the Balkan Peninsula and almost half of its total area is agricultural land. Although, there are few economic important sectors, agriculture and food production are very significant for the state economy. The unproperly implementation of our laws in practice brings many opportunities for water and soil contamination in our country. They usually contain different elements, which can be toxic or affect the plants metabolism. Therefore, inspection on trace metals, especially heavy metals in the environment and plants are important process. Microelements present as essential metals in plants influence the synthetic pathways and the yield on primary and secondary plant metabolites important for the economic sector.

As a team, who deals with medicinal plants, having experience in electrochemical techniques we could take a part in analysis of the metals on plant metabolites. Cyclic voltammetry or square wave voltammetry are reliable and suitable for qualitative and quantitative studying of the trace metals but also the kinetic of their interaction with proteins and enzymes.

Our recent publications are giving additional theoretical modeling of the kinetic of different types of interaction of the metals and proteins. Participation in WG2 or WG3 in the PlantMetals COST Action could joined our previous interests and research aims to prospective collaboration for improvement of agricultural sector and environmental protection.

EPR spectroscopy as a Tool to Investigate PLANTMETALS

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Common essential trace metals for all plants are Cu, Fe, Mn, Mo, Ni, Zn, Cd and Pb. As micronutrients they occur ranging from deficit to toxic levels with respect to natural and anthropogenic causes and different concentrations in different environments. Electron paramagnetic resonance (EPR) spectroscopy is unique direct and non-invasive technique for detection, identification, quantification and characterization of plant system based on monitoring interactions of unpaired electrons. In aforementioned metals the unpaired electrons generally originate from the d orbital manifold. Therefore, from EPR spectrum can be derived important information about system. Conceptually the simplest information, but by no means insignificant, from g-value, is identification of the type of metal in sample, followed with providing a quantitative measure of amount of metal centres present. The second goal is to obtain insights into structural domain of unpaired electron/s found on metal centre and to characterize the local environment by probing the interaction of local electric and magnetic fields within the metal complexes. The further objective on the EPR technique is to provide some insights into the important physical processes such as redox potential, uptake, delivery and release, kinetics etc. The value of EPR techniques, "standard" (continuous-wave) and "advanced" (pulsed, high-field, double resonance, etc.) lies in providing additional information to a myriad of interesting problems.

# Iron speciation in seeds of Arabidopsis thaliana and dynamics of vacuolar remobilization during germination

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After germination, during the early stages of growth, the nutrition of the seedling is transiently sustained by elements stored in the seed. Therefore, the remobilization process is extremely important for the fate of the future plant. We have focused on the mechanistic dissection of Fe remobilization in Arabidopsis that store Fe in vacuoles of embryonic endodermal cells. First, using Synchrotron Radiation X-Ray Fluorescence and Absorption Spectroscopy, we have established that Fe is stored as Fe3+-phytate complexes. We have then searched for phytases that would be involved in the degradation of phytate and in the release of free Fe. By a candidate gene approach, we have identified three purple acid phosphatases (PAP) that participate in the phytate hydrolysis and Fe release. Knock out mutants of these PAP genes display delayed phytate degradation, slower Fe remobilization and higher sensitivity to both Fe and phosphate deficiencies. Beside this approach, a genetic screen has been developed, based on Fe imaging with the Perls/DAB histochemical procedure. Indeed, the kinetics of Fe remobilization can be monitored at the whole seedling level with the Perls/DAB. With this technique, we have highlighted some variability among Arabidopsis accessions, both in the speed and in the vascular patterning of Fe. We are thus conducting genome wide association studies (GWAS) to identify new actors of the Fe remobilization process and the preliminary results will be presented.

Foliar Si and Se supply, alone and combined with organic soil amendments, to promote crop production on metal(loid)-contaminated soils

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Field trials are established with non-food crops and grassland intended for the production of biomass at two metal(loid)-contaminated sites nearby Bordeaux, France. The influence of organic amendments (i.e. compost, greenwaste compost, and biochar) and foliar Si and Se fertilization on crop yield and the limitation of metal(loid) uptake by plants is assessed. Datasets are presented for tobacco and winter barley at a Cu/PAH-contaminated site and for grassland at a site contaminated by metal(loid)s and organic xenobiotics. For instance, compost incorporation into the soil combined with liming and foliar Si and Se supply result in lowest shoot Cu concentration in tobacco plants. Shoot Se concentration of tobacco was enhanced by foliar Se supply. Influence of foliar Si and Se fertilization, alone and combined with organic soil amendments on crop yield and metal(loid) uptake is discussed.

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The UMR BIOGECO is member of the INRAE ecotoxicology network (ECOTOX, https://www6.inrae.fr/ecotox_eng/) and the COST Action 19116 PlantMetals (https://plantmetals.eu/plantmetals-home.html). The City of Bordeaux (Bordeaux Metropole) and Lyonnet SA have given access to the sites.

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What X-rays can tell about metals in plants: localization and chemistry. Current and future work.

The fate of metals in plant tissues and environment can be analyzed with X-rays. Using benchtop micro X-ray fluorescence it is possible to study metal distribution in plant roots, whole leaves and in vivo (air and water flows). Recent advances in synchrotron sources and detection allow collecting tomography images at the micron and nanometer range allowing to study element distribution in tissues and cells down to the organelle level in the best case. Most recent X-ray optics (CRL lenses) allow using the high yield K edges of heavy metals like cadmium. MicroXANES tomography reveals the metal speciation giving insight into the uptake, transport and metal sequestration. Recent examples are: Zn accumulation and speciation in the non-hyperaccumulator Noccaea ochroleucum, the interaction of the pathogen Phomopsis longicolla with metals in soybean roots, the fate of La in Desmodesmus quadricauda, the Fe and Zn distribution in nodules of Medicago truncatula, the effect of the fungus Serendipita indica on metals in barley roots, and the Cd binding to light harvesting complexes in soybean. The examples show the advantages and limitations of the methods and provide an overview of the applications of these techniques in the field of this COST Action.

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Bioavailability, environmental and human health risk of potentially toxic elements in the soil-grapevine system

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The first step in obtaining safe grapevine production is monitoring of potentially toxic element (PTE) concentrations in vineyard samples. Moreover, assessing mobility and bioavailability is crucial for better understanding the element transportation chain. In this abstract, we investigated PTE in soil and grapevine and their influence on human health. Concentrations of Al, As, B, Ba, Be, Ca, Cd, Co, Cr, Cu, Fe, Ca, K, Mg, Mn, Mo, Na, Ni, P, Pb, S, Si, Sb, Sr, V, Zn were determined in soil, soil bioavailable fractions, grapevine, gastrointestinal extracts of soil and grapevine samples. These investigations were conducted to assess the element mobility and bioavailability in the soil−plant system and examine environmental implications, human health risk and bioaccessibility of PTE in human gastrointestinal tract. Benefits of these investigations contribute to a better understanding of PTE behaviour in the soil−plant system and to making a more representative selection of single extraction procedure for PTE bioavailability assessment. Environmental risk assessments pointed out the most polluted locations, and human health risk assessments showed grape safety. PTE bioaccessibility was assessed through in vitro models for simulating elements resorption in human gastrointestinal tract.

Bioaccumulation of potentially toxic elements in Noccaea kovatsii and N. praecox (Brassicaceae) from different geological substrates and their physiological responses to Ni

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Noccaea praecox is one of the well-known model systems in the studies of PTE accumulation and tolerance, but high metal concentrations were also observed in N. kovatsii. The potential of these two pseudometallophytes to hyperaccumulate PTE was investigated at 30 ultramafic and non-ultramafic sites on the Balkan Peninsula. Tolerance to nickel and physiological responses of these taxa were also studied. Selected populations of both species were exposed to a Ni gradient in the substrate (1000, 2000 i 4000 mg kg-1) for 3 months under controlled conditions. Ni concentrations, biomass yield, concentrations of photosynthetic pigments and various phenolic compounds in response to Ni were analyzed. Nickel hyperaccumulation was observed in all ultramafic accessions of both species, with the highest concentration of 11100 mg kg-1 in shoot samples of N. praecox from Mt Tara, while the accession of N. kovatsii from schist soil on Mt Kopaonik hyperaccumulated Zn (4920 mg kg-1), with pronounced Cd accumulation as well. Lower biomass of control plants was observed in ultramafic populations compared to non-ultramafic populations, but also with higher Ni tolerance rate. Exposure to Ni reduced chlorophyll content in most non-ultramafic populations, but content was mostly unaffected in populations from ultramafic sites. The response of phenolic content in relation to Ni concentrations vary greatly in the studied populations of both species

# Beneficial role of zinc against biotic stress in metal non-hyperaccumulating plants

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Zinc is essential for functioning of numerous proteins in plants, which are involved in plant development and environment sensing. In Zn hyperaccumulating plants, both direct toxicity (elemental defence) and synergistic effects of high Zn and organic defence (joint effects) against biotic stress have been proposed. The aim of our research is to reveal the role of Zn in defence against pathogenic fungi and viruses in non-hyperaccumulating plants including crops.
We investigated the effects of Zn supply, from deficiency to toxicity, on Glycine max root defence responses to the pathogen Phomopsis longicolla. In vivo µXRF analyses showed local Zn mobilization in the infected roots, except root tips [1]. About 3-fold above-optimal Zn supply could maintain infected-plant performance by jasmonoyl-L-isoleucine and phenolics accumulation, while toxic Zn and pathogen synergistically inhibited photosynthesis and growth. Furthermore, the role of Zn against chloroplastic turnip yellow mosaic virus in Zn-tolerant Noccaea ochroleucum was shown [2]. Adequate Zn supply and cellular distribution was essential for maintaining photosynthesis under virus infection, achieved by tissue-specific activation of metal transporter gene expression (HMA3-mesophyll; MTP1-epidermis).

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# The PlantaSYST project: establishment of a new Center-of-Excellence in Bulgaria for the translation of fundamental research into sustainable bio-based technologies

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PlantaSYST is a H2020 Teaming action that resulted in the establishment of the Center of Plant Systems Biology and Biotechnology (CPSBB) in Plovdiv, Bulgaria, with the mutual efforts of 5 institutions – 3 Bulgarian and 2 German ones. CPSBB was registered in 2015 as an autonomous research entity and is fully supported by the Bulgarian and German PlantaSYST partners, the Governments of Bulgaria and Germany, and Plovdiv Municipality. The aim of the project is to position the Center as a leading research organization in Bulgaria and South-East Europe. Soon, the Center will be situated in a newly-built campus in Plovdiv, boasting state-of-the art research infrastructure, a training corps, and 2 large high-tech greenhouses. CPSBB implements cutting-edge genetics and –omics technologies in order to unravel the plant biochemical machinery and translate this scientific knowledge into applications and products for the market. One of its departments dedicated to fundamental research is Molecular stress physiology. The work in this unit focuses on analysis of the molecular responses of crops and model plants to various abiotic stresses, including heavy metals, and on improving the performance of crops under such conditions.

# Plant metal transporters and plant-microorganisms interactions within the rhizosphere for heavy metal remediation and biofortification purposes

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Metals are essential for all forms of life, however, when present at high concentrations these metals become toxic alongside with other metal(loid)s that have no known biological function. We focus our research on (1) Remediation of heavy metal contaminated area and reducing the risks to human health from exposure to heavy metals. We are interested in identifying native metallophyte species within the spontaneous flora growing on metalliferous substrates and understand their molecular mechanism of resistance/tolerance. For maintaining the cellular metal homeostasis, plants often employ transporters. The ubiquitous CDFs (Cation Diffusion Facilitators) known in plants as MTPs (Metal Tolerance Proteins) are recognized to play a major role in the efflux of transition metal cations from the cytoplasm to the outside of the cell or into subcellular compartments. We are interested in structure-function analysis of plant MTP transporters and in manipulating them for remediation purpose. Within their rhizosphere, plants interact with numerous microorganisms (bacteria and fungi) that can aid plant growth and development especially under stress conditions such as the heavy metal contamination. We are interested in identifying suitable microorganisms (bacteria and fungi) that can assist plant growth for phytoremediation purposes. (2) Genetic biofortification of crops through manipulating plant metal transporters such as MTPs in order to address zinc deficiency in humans.

Acknolegments. The research is funded by the Romanian National Authority for Scientific Research and Innovation, CNCS—UEFISCDI, project number PN-III-P2-2.1-PED-2019-5254, contract no. 390PED/2020. Travel costs are supported by COST Action 19116.

Contribution of the UniPd partner to the activities of the PLANTMETAL COST Action

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The UniPd (DAFNAE) perform research in all sectors of the agri-food sciences and technologies. Within PLANTMETAL it performs research activities on speciation and bioavailability of metals and metalloids in plant rhizosphere, physiological and biochemical responses of plants to metals and metalloids, remediation of metal contaminated soil and water, dynamics of metal and metalloids in conventional and organic horticulture in long-term field experiments.
Current research activities:
1) Analysis of metal speciation in the rhizosphere and modelling of metal uptake by agricultural plants in conventional and organic farming
2) Role of plant growth promoting microorganisms in dynamics and plant uptake of metals and metalloids in the rhizosphere
3) Proteomic responses of plants exposed to metals and metalloids, and sub-cellular localization of metals and to metals and metalloids
4) Thallium availability and accumulation in horticultural plants in a Tl contaminated environment and agricultural biotechnologies for preventing potentially toxic metals and metalloids.
The Toniolo Experimental Farm of the DAFNAE runs the among the longest field trials in Europe on organic horticulture (2005) and conventional maize rotation (1964), greenhouse experiments on light quality, vertical farming and mixed agriculture, Research is also performed on the cellular and sub-cellular localization of metals and metalloids, and the related genetic and biochemical regulatory mechanisms.

Processes controlling metals in soil-plant systems: Interest of combining speciation and stable isotopes fractionation

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The transfer of metals into plants and their translocation within plants has important implications for food safety and food quality in the case of crop plants, for the management of contaminated soils. X-ray absorption spectroscopy and stable isotope geochemistry are complementary approaches to study the fate of metals in soil-plant systems since they provide a snapshot on the metal speciation in a given plant organ, whereas metal isotope ratios result from chemical and biological processes all along the pathway from the soil to the plant organ.
Three case studies will be presented, on zinc and cadmium. The first one focuses on the fate of Zn in a contaminated soil-plant system: aquatic plants, Phalaris arundinacea and Typha latifolia, growing in an urban wetland. The second one focuses on the fate of Cd in a model crop, rice that grew in soil spiked with Cd. In that case, the effect of the water management and of the root vacuolar transporter HMA3 were studied. For the third case study, first results from an ongoing project on Cd in a cocoa plantation with background Cd levels will be presented. The contrasted behavior of Zn and Cd in terms of isotope fractionation and their distribution within the plants will be discussed in light of their affinity for O and S ligands and putative metal transporters involved. Finally, the interest and limitations of these approaches will be discussed.

Acknowledgements: This research was supported by The CNRS/INSU/EC2CO program, and by the Swiss national research foundation (P2EZP2-178618 to M.W.). ISTerre is part of Labex OSUG@2020 (Investissements d'avenirdANR10 LABX56). We thank the staff of the analytical platforms at ISTerre and ENS Lyon for their support and the synchrotrons SOLEIL, ALBA, DESY and ESRF are acknowledged for the provision of beamtime, the staff of the beamlines Samba, CLAESS, P64 and FAME, and CALIPSOplus for travel reimbursement at DESY. We also thank the EU through the COST action 19116 "PLANTMETALS" for supporting this kick off meeting
.

Anticancer potential of lettuce biofortified with iodine in human gastrointestinal cancer cell lines as well as aspects of plant-derived thyroid hormone analogs in lettuce

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The objective of this study was to determine the effect of iodine biofortification KIO3, 5-iodosalicylic acid/5-ISA/, and 3,5-diiodosalicylic acid/3,5-diISA/) on the antioxidant activity of lettuce. 5-ISA and 3,5-diISA were naturally synthesized in plants, similarly to other organic iodine metabolites i.e. iodotyrosine, as well as plant-derived thyroid hormone analogs (PDTHA), that is, triiodothyronine (T3) and thyroxine (T4). T3 and T4 were synthesized in roots with the participation of endogenous and exogenous 5-ISA and 3,5-diISA and then transported to leaves. The level of plant enrichment in iodine was safe for consumers. We also evaluated the impact of extracts from iodine-biofortified lettuce on production of reactive oxygen species (ROS) in gastrointestinal cancer cells. We demonstrated that application of iodine compounds improves the antioxidant potential of lettuce by increasing the concentration of some vitamins, antioxidant enzymes and polyphenolic compounds in the enriched plants. The results from this research indicate that iodine-biofortified lettuce induces production of ROS in cancer cells, resulting in an anticancer effect by the induction of programmed cancer cell death.

# The elements content in Cedrus atlantica (Endl.) Manetti ex Carrière (Pinaceae) needles and soil of the urban forests in Belgrade (Serbia)

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Due to their worldwide distribution and the ease of sample collection conifer needles are widely used as bio-ndicators. The conifer needles are able to accumulate, throughout years, a wide range of organic and inorganic pollutants. In this study Cedrus atlantica needles were used as a biomonitor to assess the concentration of forty-eight elements in urban forests. The samples were collected in the city of Belgrade, Serbia, from fifteen sampling sites in three urban forests: Byford's and Zvezdara forest, and Avala mountain. The concentrations of forty-eight elements were measured using an X-Ray Fluorescence (XRF). To assess the concentration of forty-eight elements in urban forests soil samples were also collected. Soil samples were collected randomly from three sites from Byford's and Zvezdara forest, and nine sites of Avala mountain. Elements source apportionment was studied employing Principal Component Analysis (PCA). The differences in metal accumulation were linked to the environmental conditions at the respective sampling site, since the needles of same species was analyzed at all locations. By comparing the elemental contents of the soils and needles at the same location, it can be clearly seen, that the foliar uptake contributes to the final elements amount as well as the root.

# A European perspective on the improvement of plant mineral micronutrient homeostasis for agriculture and human nutrition

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Mineral micronutrient deficiencies are a widespread and growing problem that affects society in the value of its crops, the well-being of its citizens, and in the need to mitigate the effects of Climate Change. Micronutrient deficiencies affect crop yield and quality in many European countries, with sharp regional differences. Mild or hidden micronutrient deficiency likely limits crop yields in much wider areas than those where obvious symptoms occur. Insufficient amount and bioavailability of micronutrients in plant-based diets represents a major reason for the high prevalence of micronutrient deficiencies in human populations, which affect brain function and immune response. In Europe, these deficiencies are a growing concern among pregnant women, children and elderly people, prone to become more severe with European diet transition towards vegetarianism. In a prospective work on the need for plant improvement within the EU project CropBooster-P, we call for awareness of the importance of micronutrients in crop production. We highlight the need for further research on mechanisms of micronutrient homeostasis and fertilization of crop plants with micronutrients to meet the challenges of developing more sustainable and nutritious crops resilient to Climate Change.

# Intellectual Property rights in scientific project: visions and reality

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New ideas and results of applied science may need to be protected if commercial usage is planned. This may be done by different ways , which may deliver different levels of protection. It's also important to take to the account whether these ideas came as the results of a project supported by the EU or local grand agencies. We will focus on the practical applications, examples and how to face the bureaucracy linked to such tasks. What would be the optimal level of the protection? What is the expect financial outcome of the IP? Where and how we should protect it? All these questions and more we will discuss.

# The effect of soil cadmium concentration on the nutritional value of Brasiliana Lettuce plants

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A pot experiment was conducted to quantify cadmium accumulation in leaves and its effect on the nutritional value of Brasiliana lettuce plants. Cadmium of 1-3 ppm in soil reduced concentrations of calcium by 28% , potassium by 39%, phosphorus by 46%, copper by 37%, magnesium by 33%, chlorophyll A by 33% and chlorophyll B by 61% in the leaves of the Brasiliana Lettuce plants. Carotene concentration was not influenced bu Cd accumulation in plants tissue. We also tested the usefulness of using chlorophyll concentration as cadmium stress indicator for the Brasiliana lettuce plants.

# Effect of pine wood biochar on immobilisation of potentially toxic elements in contaminated soil and uptake by oat (Avena Sativa L.)

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Immobilisation of potentially toxic elements (PTEs) in polluted soil due to biochar addition reduces direct PTEs toxicity and leaching fraction. Oat is pollutant-accumulating plant which is suitable for the PTEs extraction in polluted soil. Hence, the current study aimed to assess PTEs (Zn, Cr, Ni, Cu, Cd, Pb) immobilisation effect of pine wood biochar incorporated into contaminated soil and PTEs uptake by oat (Avena sativa L.). Pine wood biochar (700°C) was incorporated into former sewage sludge soil at three ratios (1/5; 1/10; 1/20) and different experiments were performed: 1) leaching; 2) bioaccumulation. According to the leaching experiment, PTEs retention efficiency in amended soil decreased in such order: Pb (99.6%) > Cu (49.3%) > Zn (37.6%) > Cr (15.2%). PTE concentrations in plants grown in biochar amended soil decreased, except in the case of Pb, which is possibly accumulating in oat due to soil acidification from 6.79 to 6.44 at the end of incubation experiment. Biochar had negative effect on the dry and fresh oat mass, which decreased by 12.1% and 33.8%, respectively.

# Alleviation of metal(loid) toxicity in plants by silicon

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Despite the non-essentiality of silicon (Si), many plants could take up large amount of this element that exceed those of other essential macronutrients like phosphorus (P) or sulphur (S). Moreover, Si was found to ameliorate the phytotoxicity induced by excess metal(loid)s whether essential (e.g. Cu, Ni, and Zn) and also non-essential (e.g. Cd, Pb, Cr, Al, As, and Sb). The Si-enhanced resistance allowing plants to cope with this type of abiotic stress has been developed at multiple levels in various plant species. One of the first defence mechanisms is Si-based restriction of root uptake and immobilization of metals and metalloids in the rhizosphere. Several studies documented the formation of insoluble complexes between Si and metal(loid)s and their storage within cell walls as a strategy that help plants to decrease available element concentration and restrict symplasmic uptake. Silicon has also been shown to influence the oxidative stress of plants. The way of acting might be by modifying the activity of various antioxidants, improving membrane stability, and acting on gene expression. The aim of this contribution is to show an overview of all currently known plant-based mechanisms related to Si supply and its involvement in amelioration of stress caused by excess metals and metalloids.

Acknowledgement: This work was supported by the Slovak Research and Development Agency, contract Nos. APVV-17-0164 and APVV SK-PL-18-0078, and by a COST Action Nr. COST CA19116.

# Common bean (Phaseolus vulgaris L.) mineral profile is negatively affected by elevated atmospheric carbon dioxide (CO2)

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Unraveling the effects and mechanisms underlying eCO2 responses of key food crops is of upmost importance to anticipate potential negative effects in human nutrition. Here, 2 bean varieties (G1378 and Logan) were grown in field conditions (BeedFACE platform) under ambient CO2 levels (control) or eCO2 (600 pm) to unravel the effects underlying eCO2 responses on biomass, mineral profile (Mg, Fe, Zn and Mn) and related gene expression.
Exposure to eCO2 resulted in a significant increase of plant biomass in G1378 (by 1.6-fold), and of full-pod biomass in both genotypes (up to 2.5-fold). In G1378, elevated atmospheric CO2 increased Mn and Fe concentrations in leaves (up to 1.4-fold) and of Mg in mature pods (by 1.2-fold). However, in Logan, eCO2 negatively affected Zn concentration in both leaves and pods (up to 0.6-fold) and of Fe in pods (by 0.6-fold), although its concentration in plant leaves increased (by 2.0-fold). Concordantly, genes involved in Fe transport (NRAMP6 and NRAMP7) were significantly downregulated in roots of Logan, which could have resulted in the lower accumulation of Fe in grains of this genotype. Contrastingly, in G1378 FER1, involved in Fe storage, was downregulated in roots and NRAMP6 upregulated in leaves, possibly contributing to the increased Fe concentration observed. In addition, Zn transporters (ZIP1 and ZIP16) were highly upregulated in Logan, most likely to counteract the decreased Zn accumulation observed in shoot tissues of this genotype.

Different strategies of Cd resistance in Arabidopsis halleri

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Metallophytes and hyperaccumulators of trace metals in particular, are case studies to understand metal homeostasis and detoxification. Cadmium is one of the most toxic trace metals for living organisms and its accumulation in the environment is recognized as a worldwide concern. Few plants have to ability to hyperaccumulate Cd. Among them, the pseudometallophyte Arabidopsis halleri is probably the most studied species up to now. While the first knowledge has been published on some populations, based on inter-species comparison, a great intraspecific variation in the way plants regulate metal content has been lately highlighted in A. halleri. Our team has already published several comparisons between A. halleri populations, highlighting for example the role of flavonoids or metal transporters in the different adaptations. Lately our team has used contrasting populations of A. halleri to explore the role of cell wall composition in metal accumulation.
The study of the diversity of resistance and detoxification mechanisms opens new avenues in the field and will be rich in collaborations inside the PLANT METALS Cost action.

# Red mud as a secondary source of scarce metals - Recovery using red microalgae

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Red mud is a by-product of the production of alumina from bauxite ore. Less than 2% of the red mud produced is currently being reused. The red mud contains a number of residual elements, some at a considerable concentration. The red microalga Galdieria sulphuraria was used to test the accumulation of scarce metals from red mud. Algal cells were cultured autotrophically and mixotrophically in a liquid medium with an alternative addition of glycerol as a source of carbon. Red mud was added into the growth medium as the acidic extract (in 10% HNO3). The growth of the cultures was monitored. The content of single scarce metals in the red mud extract and the biomass, was determined using ICP-MS. The most abundant element in red mud was Fe followed by Na and Al (53%, 17% and 12% respectively). The most abundant lanthanides were Ce, Y and La. The growth of cultures grown in the presence of red mud was comparable with the control. The red alga Galdieria sulphuraria can grow in the presence of red mud and accumulate scarce metals from it. The accumulation is more effective under the mixotrophic regime, shoing Y as the most accumulated lanthanide.

## Venue

**On site**

The meeting will be hosted at our Biology Centre (BC) of the Czech Academy of Sciences (CAS) in České Budějovice, Czech Republic. The Biology Centre is located at the western part of the joined campus of the Czech Academy of Sciences and the University of South Bohemia.

Due to a recent severe weather event that has torn off part of the copper roof from the main lecture hall of the BC where we wanted to have the lectures, posters and workgroup meeting spaces, we moved to the building of the Faculty of Arts of the University of South Bohemia (USB), which is on the same campus (see the map at the end of this programme). The lectures will be in lecture hall P1 of this building, the posters and the photosynthesis workshop in the neighbouring seminar rooms. For the welcome party and the daily lunches we will use the BC/USB campus facilities as shown on the map provided in the invitation and at the end of this programme. In the BC/USB campus, there is public high-speed (10GBit/s) WiFi accessible via the international "eduroam" system – please obtain credentials for eduroam from your local employer (if offered). For those who don't have eduroam, we will provide access to the Wifi temporarily during the conference days.

**GPS**

DD – decimal degrees: 48.9783207 N, 14.4477556 E

DMS – degrees, minutes, seconds: N 48° 58' 41.955' E 14° 26' 51.92''

**Venue – limited access to the meeting online**

Generally, this meeting is meant to be physical, not online or "hybrid", as decided in MC1! However, as the COVIS-19 pandemic still restricts travel for some of us, we will make the lectures on 25$^{th}$ of August available for listening via Zoom (details will be sent by e-mail). We will also try to make participation in the lectures via Zoom interactive, as well as providing access to the WG discussions on Friday 27$^{th}$ in the same way. However, depending on your local internet connection there may be bandwidth problems with so many people, so that we have to decide on short notice (also depending on how many try to join that way) whether an interactive Zoom participation will be possible, or only a limited listening mode. The MC meeting will only be for physically present members to avoid any problems with access control and voting.

## International travel

České Budějovice is a large regional town (about 95 000 inhabitants) and the capital of the Region of South Bohemia (Jihocesky kraj). It is easy to reach by ground transportation as described in several variants below.

**Train**

From most neighbour countries, the most convenient and cheapest way of reaching České Budějovice is by train, using the European high-speed railway system until Prague main station (Praha hlavní nádraží = Praha hl. n.) or Linz and then a direct train from these stations to České Budějovice (which is in the middle of the train line connecting Prague to Linz). Travel times vary from the Express trains that take around 2 hours to the regular trains that take about 3 hours. Furthermore, the Express

trains have onboard highspeed WiFi and offer some snacks and drinks while they cost the same as the regular trains.

**Flying**

Prague Airport Vaclav Havel (PRG).

Most airlines fly to Prague, although not always are direct flights available at suitable times.

From the airport, there are regular Airport Express Bus connections to Prague main railway station (60 CZK per person, please have cash ready!). Make sure you go to the Prague main train station (Hlavní nádraží); Prague has four larger railway stations and several smaller stations. Travelling time by bus is about 35-40 minutes; a taxi is usually not much quicker but much more expensive. Trains for České Budějovice leave quite regularly. Beware, the last express train to České Budějovice leaves at 21:58. Travel times vary from the Express trains that take around 2 hours to the regular trains that take about 3 hours. Furthermore, the Express trains have onboard highspeed WiFi and offer some snacks and drinks while they cost the same as the regular trains.

Alternatively, the RegioJet busses also stop regularly at the airport, and they connect directly to RegioJet busses to České Budějovice railroad station. It takes 45 minutes to the Prague RegioJet hub, then a 45 min wait, followed by a 2:15 journey to České Budějovice. Most of these busses have on-board WiFi, WC, and individual screens for films etc. The last RegioJet bus leaves the airport at 17:30. You have to reserve tickets in advance.

Although Prague Airport (PRG) seems the most logical choice, nearby airports include the Austrian airports of Linz (LNZ, 90 km) and Vienna (VIE, 215 km). Both have regular rail connections to České Budějovice. We would not recommend Brno Turany Airport (BRQ), because connections to České Budějovice are slow.

**Private car**

When travelling with several people and not too far a distance, time- and money-wise a shared car can be an interesting alternative. České Budějovice is easy to reach, although coming from Austria or Bavaria the last section in the Czech Republic will be mainly good quality national roads (“highways”) with only short sections of motorways. Coming from the north, there are mostly motorways, but beware of traffic jams in particular around Prague.

**Travel restrictions caused by the COVID-19 pandemic**

At the time of writing this programme, still various travel restrictions related to the COVID-19 pandemic are in place. Please inform yourself regularly and well ahead of the travel at the embassy of your country about the current rules for travelling to Czechia – which tests are required before travel and whether quarantine is required. If you are already at least two weeks past a full, certified vaccination against COVID-19, you can freely enter Czechia without test or quarantine.

**Visa**

If you are from within the EU, no visa is needed. If you are from outside the EU, you might need a visa. Please check this with the Czech Embassy in your country. You have to do this early; this process normally cost time and you might need original confirmation of participation from us. In addition, still ongoing COVID restrictions might make this process even longer.

## Accommodation

Neither the University nor the Academy has suitable on-campus accommodation. However, there are numerous possibilities nearby and in the town. The best is to check with booking.com or similar online booking websites. If you prefer smaller “bed and breakfast” pensions, which are usually not listed in booking.com and similar websites, you can find them on www.mapy.cz by typing in Ceske Budejovice and then zooming in using the “+” button on the upper right corner until close to the maximal zoom level. You will see that with each zoom level more of the accommodation symbols appearing – there are very many of those in town, usually they are good (sometimes excellent) quality and inexpensive. www.mapy.cz always displays the webpages or phone number of these pensions when you click on them - not all but many of the owners speak English or German! We recommend to book as soon as possible, because on Thursday August 26 the very popular national agricultural exhibition “The providing Earth” starts, and popular accommodation will be booked out soon.

## Local travel·

**Walking**

The BC is about 2 km from the historical centre of the town, to the main square it is 2.3 km on foot (36 min according to mapy.cz) via the shortest route or 2.8km (43 min) via a more scenic route along the “Stromovka” park.

Alternatively, you can use the local public transport; several lines regularly pass the BC (stopping point University):

**Autobus and Trolleybus**

These two lines are going from the railway station to the western outskirts, passing the university stop. You can join this bus just a short walk to outside the historic center ring. The main connection is Line 3 to Máj, Antonína Barcala (to Nadrazi if going to town). This goes very frequently from early morning until late evening. Alternatively, or additionally depending on the location of your accommodation, you can also take lines 7, 15, or 45 and some long-distance buses to/from BC. Your accommodation provider can give you more information on this, or you can search for connections on https://www.seznam.cz/jizdnirady/.

**For coming to BC from town, private cars are NOT recommended due to limited parking space.**

# *Editorial*

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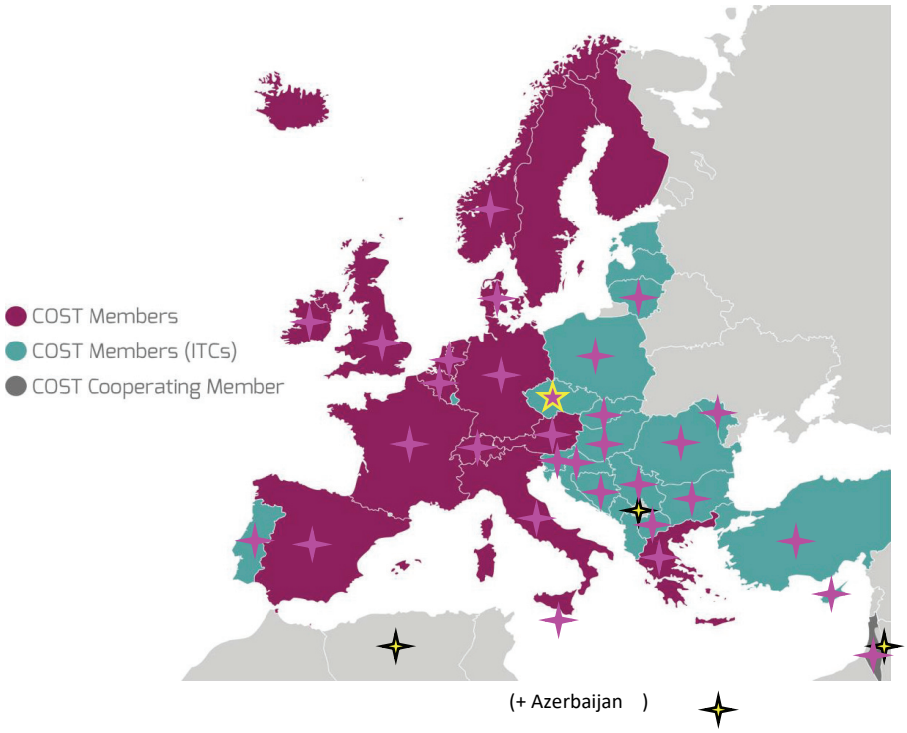
**map showing the participating countries in this COST Action**

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BIOLOGY CENTRE ASCR

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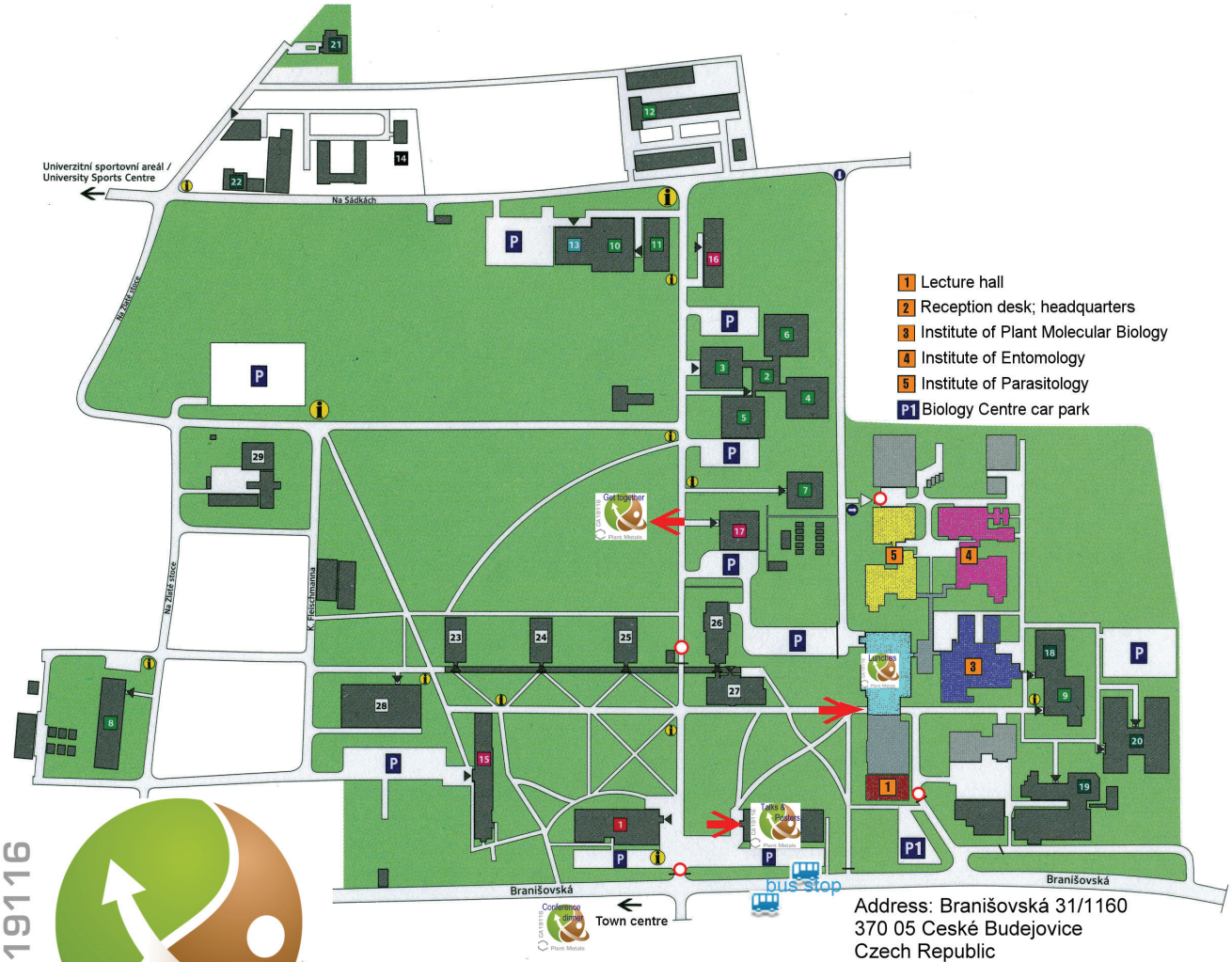
Jihočeská univerzita v Českých Budějovicích
University of South Bohemia in České Budějovice

→ way to the locations of the PLANTMETALS kick-off meeting

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Czech Republic

- **Rektorát / Rector's Office**
- 1 Akademická knihovna / Academic Library
- **Filozofická fakulta / Faculty of Philosophy**
- **Zemědělská fakulta / Faculty of Agriculture**
- 2 ZF – budova A, studijní oddělení / Building A, Student Affairs Office
- 3 ZF – budova B, katedry / Building B, Departments
- 4 ZF – budova C, centrum agroekologie / Building C, Centre for Agroecology
- 5 ZF – budova D, centrum zpracování produktů / Building D, Centre for Agricultural Product Processing
- 6 ZF – budova E, centrum hodnocení kvality / Building E, Quality Control Centre
- 7 ZF – budova M, batcentrum, učebny / Building M, BAT Centre, Classrooms
- 8 ZF – budova K 200, učebny / Building K 200, Classrooms
- 9 ZF – biologické centrum, učebny / Biology Centre, Classrooms
- 10 ZF – učebny, laboratoře / Classrooms, Laboratories
- 11 ZF – budova údržby / Service Building
- 12 ZF – sklady / Stock Rooms
- **Fakulta rybářství a ochrany vod / Faculty of Fisheries and Protection of Waters**
- 13 FROV – ústav akvakultury, ředitelství, učebny / Department of Aquaculture, Directorate, Classrooms
- 14 Školní zemědělský podnik / Agricultural School Farm
- Biologické centrum AV ČR / Biology Centre AS CR
- **Ekonomická fakulta / Faculty of Economics**
- 15 EF – budova A / Building A
- 16 EF – budova C / Building C
- 17 EF – budova F / Building F
- **Přírodovědecká fakulta / Faculty of Science**
- 18 PřF – budova A / Building A
- 19 PřF – budova B – Blažkův pavilon / Building B – Blazek Building
- 20 PřF – budova C / Building C
- 21 PřF – botanická vila / Department of Botany
- 22 PřF – centrum polární ekologie / Centre for Polar Ecology
- 23 Kolej K1 / Dormitory K1
- 24 Kolej K2 / Dormitory K2
- 25 Kolej K3 / Dormitory K3
- 26 Kolej K4 / Dormitory K4
- 27 Aula, hostel / Auditorium, Hostel
- 28 Menza / University Canteen
- 29 Vědeckotechnický park / Science and Technology Park

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Plant Metals

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MINISTRY OF EDUCATION, YOUTH AND SPORTS