

10th

INTERNATIONAL CONFERENCE ON SERPENTINE ECOLOGY

12-16 JUNE 2023 NANCY, FRANCE



ICSE
2023
NANCY, FRANCE



CONFERENCE GUIDE

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Welcome to ICSE 2023!

We are pleased to welcome you to the 10th International Conference on Serpentine Ecology in Nancy, France on 12–16 June 2023.

The International Conference on Serpentine Ecology (ICSE), directed by The International Serpentine Ecology Society (ISES), has been held every three years since 1991.

It is one of the major international scientific forums in the field of serpentine ecology, bringing together botanists, zoologists, microbiologists, physiologists, geneticists, geologists, soil scientists, and other applied specialists studying the ecology of ultramafic rocks and soil.

A notable aspect of the ICSE is the multidisciplinary nature of research on ultramafic biota, including diversity, ecology, evolution, physiology, and applied research in phytotechnologies and conservation. The main goal of the ICSE is to create a platform for the exchange of ideas and experiences, and to promote scientific dialogue, among scientists from numerous fields who share interests in the study of ultramafic habitats worldwide.

*Guillaume Echevarria
Antony Van der Ent*



HISTORY OF ICSE'S

Since 1991, researchers from approximately 45 nations have participated in nine International Conferences on Serpentine Ecology (ICSE). Conference delegates have come from all corners of the world, including Albania, Australia, Bulgaria, Canada, China, Cuba, Czech Republic, DR Congo, France, Germany, Greece, India, Iran, Italy, Japan, South Korea, New Caledonia, New Zealand, Portugal, Russia, South Africa, Spain, Sri Lanka, UK, and USA, among others. The ICSE conferences are coordinated by the International Serpentine Ecology Society (ISES), a formal research society whose members study geological, pedological, biological, and applied aspects of ultramafic ecosystems worldwide. Each conference has highlighted a region with intriguing ultramafic soil-biota relations.

- 1st ICSE in Davis, California, 1991
- 2nd ICSE in Nouméa, New Caledonia, 1995
- 3rd ICSE in Kruger National Park, South Africa, 1999
- 4th ICSE in Havana, Cuba, 2003
- 5th ICSE in Siena, Italy, 2006
- 6th ICSE in Maine, United States 2008
- 7th ICSE in Coimbra, Portugal, 2011
- 8th ICSE in Sabah, Malaysia, 2014
- 9th ICSE in Tirana, Albania, 2017
- 10th ICSE in Nancy, France, 2023

These conferences have provided an international forum to discuss and synthesize multidisciplinary research and have provided opportunities for scientists in distinct fields and from different regions of the world to conduct collaborative and interdisciplinary research. The 8th ICSE was hosted by Sabah Parks in Malaysia, on the island of Borneo, and attracted the largest delegation to date, 174 participants from 31 countries. The ninth edition took place in Albania in 2017.

Progep'Events accompanied you in 2017, and will also accompany you for this edition in 2023, we simply changed our name. Progep'Events has become UL Propuls!



9th International Conference on Serpentine Ecology

ville de Nancy,

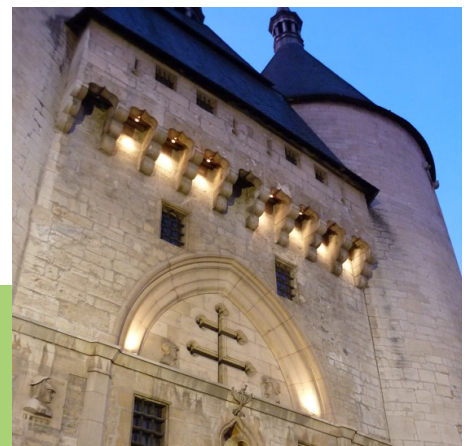
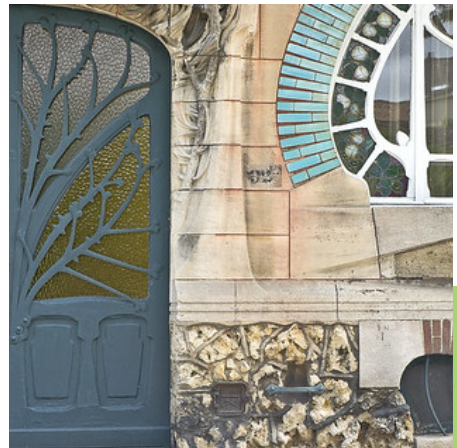
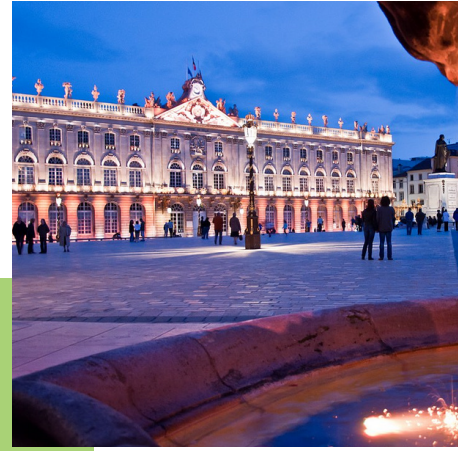
The city of Nancy is delighted to welcome you for the 10th International Conference on Serpentine Ecology (ICSE 2023).

Nancy is located in North-East of France, in the heart of Europe, not far from Luxembourg, Germany and Belgium. Nancy is a city of Art and Science. With its 18th century UNESCO World Heritage Sites, its rich “Art Nouveau” heritage, outdoor terraces and relaxed way of life, you are guaranteed to fall in love with our city.

The “Place Stanislas” has been on the UNESCO World Heritage list since 1983 and is one of the most beautiful squares of the world. Famous Place Stanislas will never fail to impress you. Its striking beauty and lively ambiance give an Italian air to the city, year round. The Place d’Alliance, which resembles a mini Place des Vosges in Paris and The Place de la Carrière also received the honour.

The artistic movement “Art Nouveau” was created in Nancy, this style celebrated the beauty of Nature in a novel decorative art. Manufactures of luxury decorative arts such as Daum or Baccarat are still present around Nancy.

Through the various social activities that will be offered throughout your stay, we hope to make you discover the different facets of the city of Nancy, between “Art Nouveau” and gourmandise.



ICSE 2023 COMMITTEES

ORGANIZING COMMITTEE

Prof Emile BENIZRI, *University of Lorraine, France*
Prof Eleonora BONIFACIO, *University of Turin, Italy*
Dr Michel CATHELINEAU, *University of Lorraine, France*
Dr Michele Eugenio D'AMICO, *University of Milan, Italy*
Dr Alexis DURAND, *University of Lorraine, France*
Prof Guillaume ECHEVARRIA, *Econick, France*
Dr Sergio Enrico FAVERO LONGO, *University of Turin, Italy*
Dr Baptiste LAUBIE, *University of Lorraine, France*
Prof Jean-Louis MOREL, *University of Lorraine, France*
Prof Marie-Odile SIMONNOT, *University of Lorraine, France*
Dr Catherine SIRGUEY, *University of Lorraine, France*
Dr Antony VAN DER ENT, *Wageningen University & Research, The Netherlands*

SCIENTIFIC COMMITTEE

Prof Alan J.M. BAKER, *School of BioSciences, The University of Melbourne, Melbourne, Australia**
Prof Aida BANI, *Agricultural University of Tirana, Albania*
Prof Robert BOYD, *Auburn University, Alabama, USA*
Prof Guillaume ECHEVARRIA, *Econick, France*
Prof Cristina GONNELLI, *University of Florence, Italy*
Dr Sandrine ISNARD, *Institute of Research for Development, France*
Dr Takafumi MIZUNO, *Mie University, Japan*
Prof Joe POLLARD, *Furman University, USA*
Prof Nishanta RAJAKARUNA, *California Polytechnic State University, San Luis Obispo, California, USA*
Prof Roger D. REEVES, *Palmerston North, New Zealand**
Dr Antony VAN DER ENT, *Wageningen University & Research, The Netherlands*

*Founding members

HOSTS & CO-HOSTS

University of Lorraine

<https://www.univ-lorraine.fr/>



As a university of all disciplines, the **University of Lorraine** encourages the sharing of knowledge. From the fundamental sciences to the humanities, it creates transdisciplinary eco-systems for innovation, which accelerate the transition from knowledge to applications. At the heart of Europe, the University of Lorraine relies on a network of partner universities in the Greater Region and throughout the world. Its international influence is based on the mobility of researchers, professors and students and on the internationalisation of its programmes.

LSE

<https://lse.univ-lorraine.fr/>



LSE (Soil and Environment Laboratory) is a research laboratory of the University of Lorraine and INRAE dedicated to the soils studies. The scientific strategy of LSE aims to answer the issues raised by the major anthropogenic pressures on soils, leading to their increasing artificialization. Are concerned, major environmental issues such as the preservation of biodiversity, the supply of biomass, or even the restoration of the quality of environments often degraded by (multi) contamination. The result is the necessary development of soil sciences in highly anthropized environments - methods and tools for the diagnostic of soil quality based on knowledge of their functioning and their evolution - and of an agronomy of highly anthropized environments - optimization of culture based on a better knowledge of the interactions between soils, plants and organisms (microflora and soil fauna). The applied part of the research carried out by LSE calls for the development of treatment processes for highly anthropized environments. These processes depend from both geosciences applied to the environment (soil engineering - soil construction) and agronomic engineering (plant engineering - phytomanagement, agromining, urban agriculture). The activities of LSE also contribute to the development of tools for taking soil into account in land planning strategies, the objective being to have functional soils able to provide ecosystem services.

ECONICK

<https://www.econick.fr/>



Econick is a plant biotechnology company producing eco-friendly metals from plants. It is aimed at any customer wishing to engage in an ethical approach to metal sourcing. The company develops unique solutions in the world making the most of the diversity and performance of certain plants to offer customized metal compounds from sustainable production processes and develop turnkey projects to enhance the value of metal-rich neglected land by establishing an economic activity that respects the environment.

INRAE

<https://www.inrae.fr/>



INRAE, the French National Research Institute for Agriculture, Food and the Environment, is the leading research organization specializing in its three scientific fields and is helping to meet these challenges. Through research, innovation and support to public policies, INRAE offers new orientations to assist the emergence of sustainable agricultural and food systems. INRAE's ambition is to provide solutions for life, humans and the earth.

ENSIC

<https://ensic.univ-lorraine.fr/>



ENSIC (National School of Chemical Industries) was founded in 1887 and is part of the Université de Lorraine in Nancy, France. As a pioneer engineering school in the fields of chemical engineering and process engineering the school aims to train engineers to rise to the challenges of the present and the future: energy, chemistry, environment, sustainable development, biotechnology, health and innovative technologies. Today there are over 4000 ENSIC engineers worldwide. Almost all of our professors at ENSIC are also researchers and work in a research laboratory on site, whether it is the LRGP (Reactions and Chemical Engineering Laboratory) or the LCPM (Macromolecular Chemistry-Physics Laboratory).

UL Propuls

<https://ul-propuls.fr/>



UL Propuls' objective is to contribute to innovation and development of territories, by helping to promote research activities (in particular by organizing scientific congresses) and by facilitating access to the know-how and companies of the know-how and equipment of the research components of the University. UL Propuls offers scientific and technical services for companies and communities, with the help of personnel and equipment of the research component of the University of Lorraine. UL Propuls also supports the promotion and the scientific of its partners, by offering them services for the offering them services for the organization of scientific events (conferences, congresses, seminars).

CONFERENCE VENUE

Access

The ICSE 2023 conference (10th International Conference Serpentine Ecology) will be held on **ENSIC Campus** (Ecole Nationale Supérieure des Industries Chimiques). ENSIC is located in the city center of Nancy, at a walking distance from the railway and tram station.

You can access the ENSIC by:

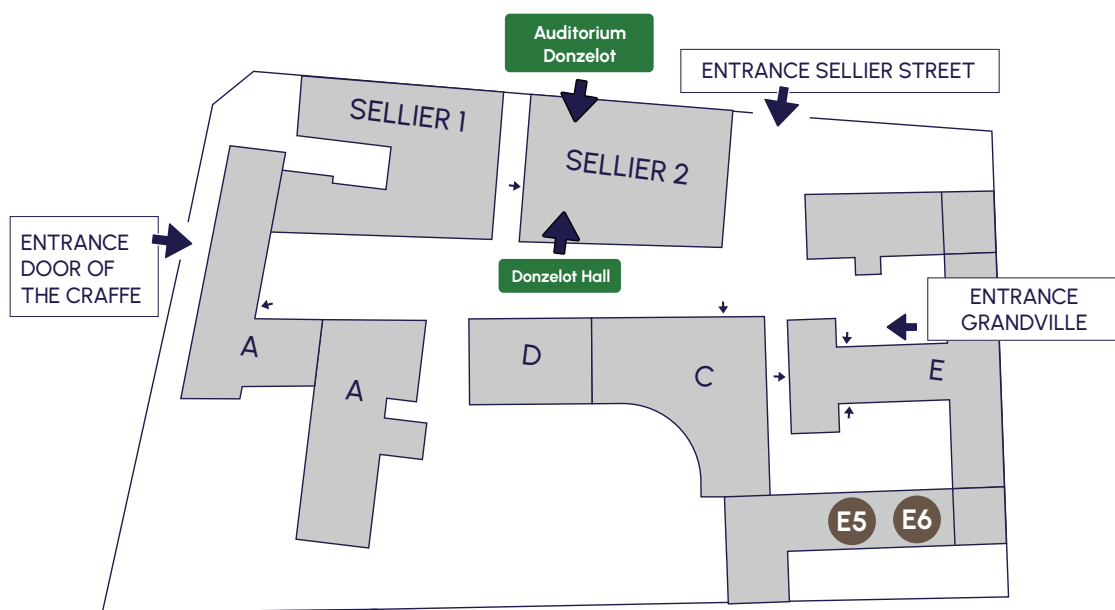
- The **entrance Grandville**, by the large wooden door (ENSIC main entrance).
- The **door of the Craffe**, street of the Citadel, by the blue grid.
- The **Sellier street**, by the blue grid.

Address

ENSIC, 1 rue Grandville, 54000 Nancy, FRANCE.

Spaces used for the conference

- **Auditorium Donzelot**: scientific sessions.
- **Donzelot Hall**: welcome desk.
- **Rooms E5 & E6**: coffee breaks, lunches and poster exhibition.



Conference badge

The ICSE conference is held on a university campus. We ask you to wear your badge for the duration of the congress, in particular to access coffee breaks, collect your lunch bag and participate in the social program.

Welcome desk

A permanence will be ensured at the reception desk for the duration of the conference, in **Donzelot Hall**, building **Sellier 2**. Do not hesitate to come to us if you want information about administrative, technical, organizational social or scientific aspects.

Welcome desk opening hours

Monday 12th: 08:00 to 16:00
Tuesday 13th: 08:00 to 12:30
Wednesday 14th: No desk (Mid tour conference)
Thursday 15th: 08:15 to 15:45
Friday 16th: 08:00 to 12:30

Organization team

The organizers of the congress will be easily identifiable, wearing a **brown scarf**. Do not hesitate to contact them for any questions.



Lunches and coffee breaks

- On Monday 12th, a buffet lunch will be held in rooms E5 & E6 (ENSIC Campus).
- On Tuesday 13th, the lunch will take place to « **Le Palais Gourmand** », which is very close to the ENSIC Campus at a walking distance.
- On Wednesday 14th, lunch bags will be provided upon presentation of your badge during the mid-conference tour.
- On Thursday 15th, the lunch will take place to « **La Petite Cuillère** », also very close to the ENSIC Campus at a walking distance.
- On Friday 16th, a lunch bag will be provided upon presentation of your badge in **Donzelot Hall**.

Morning coffee break	Lunch	Afternoon coffee break
Monday: 10:30 - 11:00 Tuesday: 10:00 - 10:30 Thursday: 10:25 - 11:00 Friday: 10:15 - 10:45	Monday: 12:15 - 13:30 Tuesday: 11:45 - 13:35 Thursday: 11:45 - 13:30 Friday: 12:15 - 13:15	Monday: 15:15 - 16:00 Tuesday: No coffee break Thursday: 15:15 - 15:45 Friday: 15:10 - 15:40

Wifi

Network	Login name	Password
eduroam	w.icse@invites	Jearwp57

Contact

icse@ul-propuls.fr
+33 (0)3 72 74 37 14

ACCESS & TRANSPORT

Nancy is a city that can be visited on foot. You are always within 5 minutes of a park or garden, your hotel, or a good restaurant.

BIKE

If you want to move quickly in **Nancy for short trips, VéloStanLib' offers bicycles free access 24/24, 7/7 and in 29 stations** particularly well distributed in the city. Many bike paths allow you to go through Nancy. It is an ecological way, simple and fast to move you in Nancy. Access the VéloStanLib' website: www.velostanlib.fr/.

BUS

Taking the bus is the quickest and cheapest way going to/coming from the city center. For a map of the bus routes and prices, **check the Stan website on www.reseau-stan.com**. We suggest you to by a ticket named "PASS10" which allowed you 10 trips (can be shared with another person).

TAXIS

Nancy taxis are available 24-hours a day, seven days a week

- Phone: +33 3 83 376 537
- Email: contact@taxis-nancy.com
- Website: www.taxis-nancy.com

LORRAINE SHUTTLES TO NANCY TRAIN STATION

- If you are travelling from Lorraine TGV station or need to get there, you can take a shuttle bus. Here are the timetables:

<https://www.keolissudlorraine.com/fiche-horaires-navette-tgv/>

- Shuttles are also available for getting to and from Lorraine airport. Access to schedules: <https://lorraineaeroport.com/passagers/acces-parking/>

Arrival and departure: Train station Nancy-St Léon.

This shuttle is available on arrival and departure from the main flights and train journey.

GALA DINNER

For the gala dinner, buses are provided to and from the venue.

Departure time from ENSIC: 18:45

Departure time from Fort Pélissier: 22:30

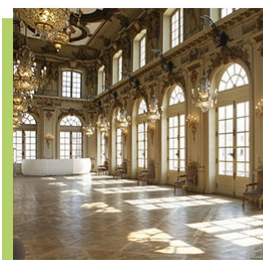
SOCIAL PROGRAM

Welcome reception

Monday, June 12th | 18:30 - 22:00 | ENSIC Campus

Included in the registration fees

You are warmly invited to the ENSIC Campus for the welcome reception. Around appetizers, refreshments and other specialties of Lorraine, enjoy this moment to exchange with the organizers and participants, to finish warmly this first day. The welcome cocktail will begin with an oral presentation by Robert Boyd in Donzelot Auditorium and will continue in rooms E5 and E6.

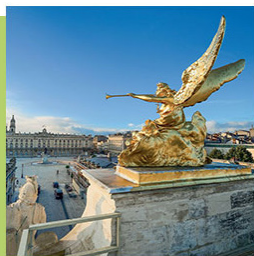


City tour

Tuesday, June 13th | 16:30 - 18:00 | Starting: ENSIC campus side Porte de la Craffe | Arrival: Place Stanislas | *Included in the registration fees*

From the origins of Nancy to the meeting in the kingdom of France under Stanislas, you will discover the emblematic sites, witnesses of the glory of the dukes of Lorraine (ducal palace, Cordeliers church, door of the Craffe, mansions ...) as well as the royal ensemble classified by UNESCO including the famous place Stanislas.

For this visit, a guide will come in front of one of the entrances to the ENSIC Campus, where the conference will take place. It will end then Place Stanislas, in full heart of the city. The visit will be organized in 3 groups of 25 people each.



SOCIAL PROGRAM

Mid-conference tour

Wednesday, June 14th | 08:00 - 18:00 | Starting: ENSIC Campus at 08:00
| Arrival: ENSIC Campus at 18:00 | Included in the registration fees, lunch bags also included

During this conference, we will propose you a mid-conference tour in to discover serpentines on the Vosges territory. You will observe the typical serpentine soils and vegetation of serpentine flora of the Vosges. You will be completely taken care of the mid-conference tour.

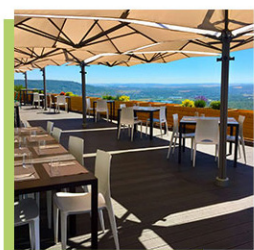
Shuttles will pick you up directly at the ENSIC campus at 08:00 and will accompany you throughout the day on the various sites. At the end of the day, you will be directly taken to the ENSIC campus. A lunch bag with a starter, a main course, a dessert and a bottle of water are included for this excursion.



Gala dinner

Tuesday, June 13th | 18:45 - 23:00 | Fort Pélissier | Meeting point: 18:45, ENSIC Campus | Included in the registration fees

We invite you to the impressive Fort Pélissier, located 15km South of Nancy, on Thursday 15 June from 19:00 to 23:00, for the gala dinner. The dinner will be served at the Auberge du Fort Pélissier restaurant. The panoramic view from the terrace and the chef's refined cuisine will awaken all your senses. During your meal, enjoy the calm and the breathtaking view in the heart of nature.



POST-TOUR CONFERENCE

From Saturday, June 17th to Tuesday, June 20th

Post-symposium excursion in ultramafic areas of North-Western Internal Alps, Valle d'Aosta and Piemonte, Italy

Departure on June, 17th to Saint Vincent (Italy)

Return on June 20th to Nancy (France)

Saturday 17 June

1st stop: Saint Vincent

Travel from Nancy to Saint Vincent. Serpentine area of the 'Roman Bridge-Tsailleun' Geosite (Saint Vincent, 560-700 m a.s.l.), with a rich xerothermic (pseudo-steppe) vegetation, including *Alyssum argenteum* and *Notholaena maranthae*, alternating with vineyards and ancient cereal fields 'suspended' between the serpentinite outcrops. In the evening, we'll reach the village of Champoluc (Val d'Ayas), at the feet of Monte Rosa where we have dinner and sleep at a/some hotels.



Sunday 18 June

2nd stop: The Verra Grande Glacier forefield (ca. 2200 m asl, Val d'Ayas)

A moraine chronosequence where soil development and vegetation succession can be seen. The limiting conditions caused by pure serpentinite are particularly visible, and even small amounts of P-richer gneiss shape soil development and primary succession in a very short time. We will see the primary vegetation succession and associated soils from pioneer communities on fresh moraine tills to subalpine forest (climax conditions), with a rich set of serpentine endemics like *Cardamine plumieri*, *Noccaea sylvia* and *Carex fimbriata*. We will spend another night in Champoluc.



Monday 19 June

3rd stop: Around the Lanzo Peridotite Massif

After leaving Champoluc in the morning after breakfast, we will exit Aosta Valley to reach the Metropolitan area of Torino. We'll visit the Riserva Naturale Madonna della Neve sul Monte Lera (1350 m; Royal Park network). This lies in the Lanzo Peridotite Massif, generated in an ocean-continent transition context and still preserving slightly serpentinized peridotites. Here, the mixed mesophilous forest develops on a lherzolite substrate and this is the sole site of the serpentine endemic *Euphorbia gibbelliana*. Soils nearby (Varisella) show an impressive development and "lateritic" crusts are common on the river banks, as evidences of past climates.



Tuesday 20 June

4th stop: Balangero

On the last day, we'll visit the northern part of the Lanzo Massif and, particularly, the abandoned chrysotile mine of Balangero and Corio (550-890 m), the former largest asbestos mine in Western Europe, where mining activities stopped in 1990. The horizontal terraces of the open mine pit still host a pioneer vegetation, including hyperaccumulators, while a remarkable plant cover developed on the huge deposits of mine tailing, which underwent to hydrogeological stabilization and revegetation interventions. After the visit, we will reach the Torino Airport or we will travel back to Nancy, we will reach in the evening.



KEYNOTES

KEYNOTE 1 - Dr. Robert BOYD

Auburn University's first Assistant Provost for Institutional Effectiveness

Day 1: Monday, June 12th - from 18:30 before the welcome reception



Dr. Robert (Bob) Boyd is Professor in the Department of Biological Sciences at Auburn University in Alabama, USA. He received his PhD in Botany from the University of California, Davis and in 1988 became Assistant Professor at Auburn University. Boyd became Professor in 2000, became the department's Undergraduate Program Officer in 2013, and served as Interim Chair for Fall Semester 2015. In July 2019 he was named Associate Dean for Academic Affairs in the College of Sciences and Mathematics and in January 2023 he became Auburn University's first Assistant Provost for Institutional Effectiveness. Boyd has taught courses ranging from freshman introductory biology to graduate-level classes. His research interests include the management of rare and endangered plants, as well as the ecology and evolution of metal hyperaccumulator plants. He has published over 100 journal articles/book chapters, has been a guest editor for special issues of three scientific journals, and has served on the editorial boards of three additional scientific journals. A PLOS Biology paper from October 2020 listed him among the world's top 2% of plant ecologists based upon his citation metrics.

KEYNOTES

KEYNOTE 2 - Prof. Nishanta RAJAKANURA

Professor in plant biology, California Polytechnic State University

Day 2: Tuesday, June 12th - from 13:35 to 14:15



Nishi Rajakaruna is a geoecologist broadly interested in how ‘harsh’ soil conditions influence plant, lichen, and microbe diversity from species to community levels. Nishi received his BA in Human Ecology from College of the Atlantic, Maine and his MS and Ph.D. in Botany from the University of British Columbia, Canada. He conducted postdoctoral research at Stanford University and was a Fulbright US Scholar in Sri Lanka/India (2016-2017) and South Africa (2022-2023). He has taught botany for 18 years and is currently a Professor of Plant Biology at California Polytechnic State University, San Luis Obispo, California. His primary area of research is on the diversity, ecology, evolution, and conservation of plants and lichens of serpentine and other ‘harsh’ soils. He has ongoing research in California, Maine, South Africa, Russia, and Sri Lanka. Nishi has published over 90 peer-reviewed papers on geoecology and is the co-editor of two key treatments on plant life on serpentine soils--Serpentine: Evolution and Ecology of a Model System and Soil and Biota of Serpentine: *A World View--and a book titled Plant Ecology and Evolution in Harsh Environments.*

KEYNOTES

KEYNOTE 3 - Prof. Roger REEVES

Former professor in chemistry, Palmerston North, New Zealand

Day 4: Thursday, June 15th - from 08:45 to 09:25



Following Bachelor's and Master's degrees in Chemistry in New Zealand and doctoral studies in Physical Chemistry in the US on the theory and practice of viscosity measurement of molten electrolytes at high temperature, Roger Reeves was appointed in 1965 to a Lectureship in Chemistry at Massey University (NZ). He soon became associated with Robert Brooks, whose background was in Analytical Chemistry, and they collaborated on many projects using spectrometric methods for trace element analysis of a variety of geological and biological materials. In 1974, a chance meeting with Tanguy Jaffré, visiting NZ from New Caledonia, led to collaborative work there, with a particular focus on several plant species found to have accumulated high nickel concentrations from the ultramafic soils.

Initially a side-line to Roger's studies in atomic spectrometric methods of analysis (AA and ICP) and their application to many types of samples, unusual examples of trace element accumulation by plants became a major research interest, leading to almost 50 years of collaboration with plant scientists from more than 20 countries. This work now accounts for more than 140 of Roger's 200 publications. Periods of overseas leave from Massey were spent in the Chemistry Departments at the Universities of Florida and Manchester, the Botany Departments of the Universities of Sheffield, Queensland and Melbourne, and at ENSAIA in Nancy. The Massey University Metallophyte Herbarium, initiated by Robert Brooks, was developed through Roger's extensive field work into a collection of 3000 plant specimens from metalliferous soils worldwide; it is now incorporated into the Herbarium of the Royal Botanic Garden, Edinburgh.

KEYNOTES

KEYNOTE 4 - Prof. Rufus CHANEY (remote)









Day 5: Friday, June 16th - from 08:30 to 09:10



Dr. Rufus L. Chaney is a Senior Research Agronomist in the Environmental Management and By-Product Utilization Laboratory of the USDA-Agricultural Research Service at Beltsville, MD, where he conducts research on the fate, food-chain transfer, and potential effects, and remediation of hazards from soil microelements. The research includes studies on 1) plant uptake of metals and translocation to edible plant tissues; considers plant-soil interactions in microelement phytoavailability; 2) speciation of metals in plants and bioavailability to animals; 3) development of hyperaccumulator crops to phytoextract and recycle metals in contaminated soils; 4) bioavailability of lead and other metals in soils, biosolids, and composts directly ingested by animals; 5) development of «Tailor-Made Composts and Biosolids» to remediate Pb, Zn, Cd, Ni and other element contaminated soils including urban gardens; and 6) potential methods to reduce food-chain transfer or toxicity of metals in organic resources and potential regulatory approaches to protect food safety and soil fertility.

Since beginning his career in 1969, Dr. Chaney has 429 published papers and 267 published abstracts on these topics. He has cooperated with the US-Environmental Protection Agency, the US-Food and Drug Administration, the Office of Management and Budget, and many States in preparing advice and regulations for utilization of biosolids and remediation of metal contaminated soils.

SCIENTIFIC SESSIONS OF THE CONFERENCE

COLOR	SCIENTIFIC SESSION	PAGE
	1. Geology, soils, & biogeochemical cycles	p. 26
	2. Biodiversity: taxonomy and systematics	p. 28
	3. Ecology and evolution	p. 29
	4. Conservation and restoration	p. 31
	5. Ecophysiology, molecular biology, and genetics	p. 32
	6. Metal hyperaccumulation discovery & monitoring	p. 34
	7. Plants of other edaphically challenging substrates	p. 36
	8. Agromining and phytotechnologies	p. 37

PRESENTATION INSTRUCTIONS

General information for all oral presentations



- Your presentation must be **in English**.
- Your presentation support saved as **PowerPoint** or **PDF format** on a USB key.
- **For speakers on the first morning (Monday, June 12th), your presentation should be sent before the conference to icse@ul-propuls.fr indicating your session, the reference of your presentation and the title of the paper.**
- If you want to use **particular formats such as video**, sending your presentation in advance **is mandatory**.
- We invite you to load your presentation before the beginning of your session, **half a day before your presentation on the computer of your conference room**.
- To avoid any technical bug and too long installation time, it is mandatory to **only use the computer at your disposal**. We ask that you do not use your personal computer (unless otherwise indicated).
- A remote control with laser pointer will be at your disposal.



Keynote

- You will have **40 minutes** for your presentation (35 minutes of presentation + 5 minutes of question).



Regular talk

- You will have **15 minutes** for your presentation (12 minutes of presentation + 3 minutes of question).



Poster

- Your poster should be **printed in A0 format** (84.1 cm x 118.9 cm) in **portrait orientation**.
- Your poster should be **in English**.
- The posters will be displayed on grids and fixed with clips, which will be given to you upon your arrival.
- It is not possible to print your poster on site.
- Thank you for **hanging your poster** the first morning of the conference, on Monday, June 12th, **as soon as you arrive at the congress**. Thus, everyone will be able to watch it during the breaks and the poster session.
- Two poster sessions are scheduled, **Monday, June 12th from 13:30 to 14:15** and **Thursday, June 15th, from 13:30 to 14:00**. During these sessions we ask you to **be present near your posters in order to present them**.
- Please **pick up your poster** on the last day of the conference, on Friday, June 16th, the remaining posters will not be retained.

SCIENTIFIC PROGRAM: OVERVIEW

CSE 2023 Detailed Scientific Program						
Time	Monday 12 th Session	Tuesday 13 th Session	Wednesday 14 th Mid tour	Thursday 15 th Session	Friday 16 th Session	
08:45	Opening ceremony <i>Donzelot auditorium</i>	Session 3: Ecology and evolution <i>Donzelot auditorium</i>	(Including lunch) Mid conference tour Meeting at 08:00 at ENSIC Back to ENSIC at 18:30	08:45	Session 8: Agromining and phytotechnologies <i>Donzelot auditorium</i>	
09:15	Session 1: Geology, soils, & biogeochemical cycles <i>456114 - Andriade</i>	448756 - Pillon			Keynote: Rufus Chaney (remote)	
09:30	447363 - Yang	450612 - Hidalgo-Triana		09:25	461039 - Babot-Kostecka	
09:45	454685 - Ansart	450555 - Takazoe		09:40	450660 - Pollard	
10:00	449584 - Vignati	448164 - Putra		09:55	450361 - Gervais-Bergeron	
10:15	454757 - Tognacchini	Coffee Break (30") <i>Rooms E5 & E6</i>		10:10	448978 - Misljenovic	
10:30	Coffee Break (30") <i>Rooms E5 & E6</i>			10:25	Coffee Break (35") <i>Rooms E5 & E6</i>	
11:00	450081 - Paul	450347 - Havlikova		11:00	450705 - Bani	
11:15	450463 - Amir	442032 - Hukshof		11:15	455380 - Guittony	
11:30	450629 - d'Amico	448633 - Dierue		11:30	456272 - Echevarria	
11:45	455457 - Kram	450615 - Sakaguchi		11:45	460650 - Chatzelli	
12:00	455458 - Tisserand	450448 - Quintela Sabaris		11:45	450838 - Alvarez-Lopez	
12:15	Lunch (1:15) <i>Rooms E5 & E6</i>	Lunch (1:45) <i>Le Palais Gourmand</i>		13:30	Poster session (30") <i>Rooms E5 & E6</i>	
13:30	Poster session (40") <i>Rooms E5 & E6</i>	Session 7: Plants of other edaphically challenging substrates <i>Donzelot auditorium</i>			Session 5: Ecophysiology, molecular biology, and genetics <i>Donzelot auditorium</i>	
14:15	Session 2: Biodiversity, taxonomy and systematics <i>Donzelot auditorium</i>	13:35		14:00	448388 - Merlot	
14:30	450674 - Bianchi	14:15		14:15	448789 - Aarts	
14:45	442825 - Arcousture	454581 - Nagasawa		14:30	450431 - Colzi	
15:00	442831 - Leglie	454853 - Siebert		14:45	450308 - Palm	
15:15	454807 - Filis	15:00		15:00	455459 - Schat	
15:30	Coffee Break (45") <i>Rooms E5 & E6</i>	15:15		15:15	Coffee Break (30") <i>Rooms E5 & E6</i>	
16:00	450470 - Igwe	15:40		15:45	446956 - Berazain Iturralde	
16:15	450618 - Hidalgo-Triana	15:40		15:45	446401 - Minuno	
16:30	End	15:40		16:00	454855 - Lin	
18:30	Retrospective by Robert Boyd & welcome cocktail (148254) <i>Donzelot auditorium for the speech and then in rooms E5&E6</i>	15:40		16:15	450207 - Alfonso González	
		15:40		16:30	450324 - Isnard	
		15:40		16:45	446258 - LY	
		15:40		17:00	End	
		15:40			Gala Dinner 19:00-23:00 meeting at ENSIC (1845)	

Session	Name
Session 1	Geology, soils, & biogeochemical cycles
Session 2	Biodiversity: taxonomy and systematics
Session 3	Ecology and evolution
Session 4	Conservation and restoration
Session 5	Ecophysiology, molecular biology, and genetics
Session 6	Metal hyperaccumulation discovery & monitoring
Session 7	Plants of other edaphically challenging substrates
Session 8	Agromining and phytotechnologies

S.1	Monday, June 12 th (09:15 – 12:15) Chair: Guillaume ECHEVARRIA	
Regular talk 1 09:15	15'	<p>A newly-forming ecosystem in a mined ultramafic environment in Barro Alto, Goiás State, Brazil</p> <p>Leide ANDRADE¹, Fabiana AQUINO¹, Eduardo OLIVEIRA-FILHO¹, Fábio REIS-JR¹, Cícero PEREIRA¹, Juaci MALAQUIAS¹, Edson SANO¹ and Guillaume ECHEVARRIA^{2,3}</p> <p>1. <i>Embrapa Cerrados, Brazil</i> 2. <i>INRAE, Laboratoire Sols et Environnement, France</i> 3. <i>University of Queensland, Sustainable Minerals Institute, Centre for Mined Land Rehabilitation, Australia</i></p>
Regular talk 2 09:30	15'	<p>Characterization of chromium-bearing minerals in incipient and highly-weathered serpentine soil profiles in Taiwan</p> <p>Chia-Yu YANG¹, Wei-Hao LEE² and Zeng-Yei HSEU¹</p> <p>1. <i>Department of Agricultural Chemistry, National Taiwan University, Taiwan</i> 2. <i>Institute of Mineral Resources Engineering, National Taipei University of Technology, Taiwan</i></p>
Regular talk 3 09:45	15'	<p>Ni way! The tricky Ni-biogeochemical cycle in soils under hyperaccumulator of New Caledonia</p> <p>Claire ANSART¹, Eric PAIDJAN², Christophe CLOQUET¹, Emmanuelle MONTARGÈS-PELLETIER³, Sandrine ISNARD⁴, Cécile QUANTIN⁵, Yann SIVRY⁶ and Farid JUILLOT⁷</p> <p>1. <i>CRPG – Université de Lorraine, France</i> 2. <i>BIOGECO INRAE– Université de Bordeaux, CNRS, France</i> 3. <i>LIEC– Université de Lorraine, CNRS, France</i> 4. <i>AMAP – Université de Montpellier, IRD, CIRAD, CNRS, INRAE, France</i> 5. <i>GEOPS– Université Paris Saclay, CNRS, France</i> 6. <i>IPGP – Sorbonne Paris Cité – Université Paris Diderot, France</i> 7. <i>IMPMC – Université Pierre et Marie Curie, CNRS, IRD, France</i></p>
Regular talk 4 10:00	15'	<p>Nickel and chromium speciation in a small ultramafic catchment: implications for elemental bioavailability to freshwater organisms</p> <p>Davide VIGNATI¹, Elsa SALLES¹, Thi Tuyen NGUYEN², Céline SIMON¹, Maximilien BEURET¹, Carole COSSU-LEGUILLE¹, Alexis GROLEAU², Yulia ERBAN KOCHERGINA³, Yann SIVRY² and Pavel KRAM³</p> <p>1. <i>LIEC, Université de Lorraine, CNRS, France</i> 2. <i>IPGP, Université Paris Cité, CNRS, France</i> 3. <i>Division of Geochemistry and Laboratories, Czech Geological Survey, Czech Republic</i></p>
Regular talk 5 10:15	15'	<p>Rhizosphere processes and nickel mobilization in the nickel hyperaccumulator <i>Odontarrhena chalcidica</i></p> <p>Sören RISSE, Markus PUSCHENREITER and Alice TOGNACCHINI</p> <p><i>Department of Forest and Soil Sciences, University of Natural Resources and Life Sciences (BOKU), Vienna, Austria</i></p>

Regular talk 6 11:00	15'	<p>Stocks and biogeochemical cycling of soil-derived nutrients in an ultramafic rain forest in New Caledonia</p> <p>Adrian PAUL¹, Sandrine ISNARD^{2,3}, Francis BREARLEY⁴, Guillaume ECHEVARRIA⁵, Alan BAKER^{1,5,6}, Peter ERSKINE¹ and Antony VAN DER ENT¹</p> <p>1. Centre for Mined Land Rehabilitation University of Queensland, Australia 2. AMAP, Université Montpellier, France 3. AMAP, Herbier de Nouvelle-Calédonie, New Caledonia 4. Department of Natural Sciences, Manchester Metropolitan University, United Kingdom 5. Laboratoire Sols et Environnement, Université de Lorraine, France 6. School of BioSciences, The University of Melbourne, Australia</p>
Regular talk 7 11:15	15'	<p>Studies on arbuscular mycorrhizal fungi in New Caledonian ultramafic soils: A synthesis</p> <p>Hamid AMIR, Yvon CAVALOC, Thomas CROSSAY, Valérie BURTET-SARRAMEGNA, and Linda GUENTAS</p> <p><i>Institut des Sciences Exactes et Appliquées (ISEA), Université de la Nouvelle-Calédonie, Noumea 98851, New Caledonia</i></p>
Regular talk 8 11:30	15'	<p>Ancient soils in non-glaciated mountain areas on peridotite: the Lanzo Ultramafic Massif example (TO, NW Italy)</p> <p>Michele E. D'AMICO¹, Sara NEGRI² and Eleonora BONIFACIO²</p> <p>1. DISAA, Università degli Studi di Milano, Italy 2. DISAFA, Università degli Studi di Torino, Italy</p>
Regular talk 9 11:45	15'	<p>Drivers of Cr and Ni Mobility in a Serpentine Catchment</p> <p>Pavel KRÁM^{1,2}, Jakub HRUŠKA^{1,2}, Davide VIGNATI³, Katherine PÉREZ RIVERA^{4,1}, Jan ČUŘÍK^{1,2}, František VESELOVSKÝ¹, Oldřich MYŠKA^{1,2}, Yulia ERBAN KOCHERGINA¹ and Filip OULEHLE^{1,2}</p> <p>1. Department of Environmental Geochemistry and Biogeochemistry, Czech Geological Survey, Czechia 2. Department of Biogeochemical and Hydrological Cycles, Global Change Research Institute ASCR, Czechia 3. LIEC, University of Lorraine, CNRS, France 4. Department of Biological Sciences, Virginia Tech, USA</p>
Regular talk 10 12:00	15'	<p>The Nickel Biogeochemical Cycle in a Tropical Hyperaccumulator Soil-Plant System</p> <p>Romane TISSERAND¹, Antony VAN DER ENT^{1,2}, Jean-Louis MOREL¹, Sukaibin Sumail³ and Guillaume ECHEVARRIA^{1,2,4}</p> <p>1. Université de Lorraine, INRA, Laboratoire Sols et Environnement, France 2. University of Queensland, SMI, Centre for Mined Land Rehabilitation, Australia 3. Sabah Parks, Kota Kinabalu, Malaysia 4. R&D Departement, Econick, France</p>

S.2	Monday, June 12 th (14:30 – 17:00) Chairs: Prof Federico SELVI & Prof Cristina GONNELLI	
Regular talk 1 14:15	15'	Additions to the lichen flora of Albania Elisabetta BIANCHI¹, Renato BENESPERI², Ilaria COLZI² <i>1. Department of Agriculture, Food, Environment and Forestry, University of Florence, Italy</i> <i>2. Department of Biology, University of Florence, Italy</i>
Regular talk 2 14:30	15'	Seed endosphere of Brassicales and Asterales hyperaccumulating plants harbor a stable core microbiome Julien ANCOUSTURE, Alexis DURAND and Emile BENIZRI <i>Laboratoire Sols et Environnement, Université de Lorraine, INRAE, France</i>
Regular talk 3 14:45	15'	Soil contamination levels of nickel imply changes in the bacterial communities associated to the rhizosphere and endosphere of <i>Odontarrhena chalcidica</i> Pierre LEGLIZE, Alexis DURAND and Emile BENIZRI <i>Laboratoire Sols et Environnement, Université de Lorraine, INRAE, France</i>
Regular talk 4 15:00	15'	Grassland flora of serpentine areas in North Pindus (Greece) Evangelos FILIS¹, Dimitrios KYRKAS^{2,3}, Nikolaos MANTZOS², Panayiotis DIMITRAKOPOULOS³, Georgios FOTIADIS⁴ and Maria KONSTANTINO¹ <i>1. Department of Agriculture, International Hellenic University, Greece</i> <i>2. Department of Agriculture, University of Ioannina, Greece</i> <i>3. Department of Environment, University of the Aegean, Greece</i> <i>4. Department of Forestry and Natural Environment Management, Agricultural University of Athens, Greece</i>
Regular talk 5 16:00	15'	Investigating the Diversity and Function of Microorganisms in Serpentine and Non-Serpentine Soils across California Alexandria IGWE and Michelle AFKHAMI <i>Department of Biology, University of Miami, United States of America</i>
Regular talk 6 16:15	15'	The checklist of serpentinophytes (serpentine restricted flora) as a tool for researching and conservation in serpentine ecosystems (southern Iberian Peninsula, Spain) Federico CASIMIRO-SORIGUER SOLANAS, Andrés V. PEREZ-LATORRE and Noelia HIDALGO-TRIANA <i>Department of Botany and Plant Physiology, University of Malaga, Spain</i>

S.3		Tuesday, June 13 th (08:45 – 12:00) Chairs: Prof Robert BOYD & Dr Takafumi MIZUNO
Regular talk 1 09:00	15'	<p>Community ionomics of a hyperdiverse shrubland: the maquis on ultramafic substrates of New Caledonia Juliette HOCEDEZ¹, Karine GOTTY¹, Vanessa HEQUET^{2,3}, Audrey LEOPOLD⁴ and Yohan PILLON¹</p> <p>1. Laboratoire des Symbioses Tropicales et Méditerranéennes (LSTM), IRD, INRAE, CIRAD, Institut Agro, Univ. Montpellier, France 2. AMAP, IRD, CIRAD, Herbier de la Nouvelle-Calédonie, New Caledonia 3. AMAP, Univ Montpellier, IRD, CIRAD, CNRS, INRAE, France 4. Institut Agronomique Néo-Calédonien (IAC), équipe SolVeg, New Caledonia</p>
Regular talk 2 09:15	15'	<p>Functional groups and traits of plants in Mediterranean ultramafic shrublands (Sierra Bermeja, Spain) Noelia HIDALGO-TRIANA¹, Andrés V. PEREZ-LATORRE¹, Aristide ADOMOU², Michael RUDNER³ and James H. THORNE⁴</p> <p>1. Department of Botany and Plant Physiology, University of Malaga, Spain 2. Department of Plant Biology, University of Abomey-Calavi, Benin 3. Faculty of Environmental Engineering, Weiheinstephan-Triesdorf, University of Applied Sciences, Germany 4. Department of Environmental Science and Policy, University of California, Davis, USA</p>
Regular talk 3 09:30	15'	<p>Parallel adaptation to ultramafic soils in <i>Solidago virgaurea</i> L. across the Japanese Archipelago Kiyoto TAKAZOE¹, Hiroaki SETOGUCHI¹, Atsushi NAGANO^{2,3}, Naoko ISHIKAWA⁴, Kenji HORIE⁵, Motomi ITO⁶ and Shota SAKAGUCHI¹</p> <p>1. Graduate School of Human and Environmental Studies, Kyoto University, Japan 2. Faculty of Agriculture, Ryukoku University, Japan 3. Institute for Advanced Biosciences, Keio University, Japan 4. Botanical garden, Osaka Metropolitan University, Japan 5. Asahikawa City Northern Wild Plants Garden, Japan 6. Graduate School of Arts and Sciences, The University of Tokyo, Japan</p>
Regular talk 4 09:45	15'	<p>The elemental defence hypothesis: Is there a 'missing piece'? Rocky PUTRA and Caroline MÜLLER Department of Chemical Ecology, Bielefeld University, Germany</p>
Regular talk 6 10:30	15'	<p>The role of metal accumulation in serpentine adaptation in the group <i>Alyssum montanum</i> s.l. Karolína HAVLÍKOVÁ¹, Sonia CELESTINI², Filip KOLÁŘ²</p> <p>1. Department of Ecology, Charles University, Faculty of Science, Prague, Czech Republic 2. Department of Botany, Charles University, Faculty of Science, Prague, Czech Republic</p>

Regular talk 7 10:45	15'	<p>Ultramafic ecosystems as a macroecological model Catherine HULSHOF¹, Marko SPASOJEVIC², Claudia GARNICA-DÍAZ³, Nishanta RAJAKARUNA^{4,5}</p> <p>1. Department of Biology, Virginia Commonwealth University, United States of America 2. Department of Evolution, Ecology, and Organismal Biology, University of California Riverside, United States of America 3. Biology Department, University of Florida Gainesville, United States of America 4. Biological Sciences Department, California Polytechnic State University, United States of America 5. Unit for Environmental Sciences and Management, North-West University, South Africa</p>
Regular talk 8 11:00	15'	<p>Above and belowground functional space of herbaceous serpentinicolous species Florian DELERUE and Richard MICHALET</p> <p>Université de Bordeaux, CNRS, Bordeaux INP, EPOC, UMR 5805, Pessac, France</p>
Regular talk 9 11:15	15'	<p>Genetic parallelism during recurrent serpentine adaptation in the Eurasian goldenrod in Japan Shota SAKAGUCHI¹, Kenji HORIE², Naoko ISHIKAWA³, Shuji SHIGENOBU⁴, Katsushi YAMAGUCHI⁴, Mitsuyasu HASEBE⁴, Ayano MIKI¹, Hiroaki SETOGUCHI¹, Shosei KUBOTA⁵, Osamu KURASHIMA⁵, Keitaro FUKUSHIMA⁶, Yoshihisa SUYAMA⁷, Shun HIROTA³ and Motomi ITO⁵</p> <p>1. Graduate School of Human and Environmental Studies, Kyoto University, Japan 2. Northern Wild Plants Garden, Asahikawa, Japan 3. Botanic Garden of Osaka Metropolitan University, Japan 4. NIBB, Japan 5. The University of Tokyo, Japan 6. Fukushima University, Japan 7. Tohoku University, Japan</p>
Regular talk 10 11:30	15'	<p>Tolerance to ultramafic soils is mediated by control of Ca:Mg ratio in <i>Santolina semidendata</i> and associated taxa Celestino QUINTELA-SABARÍS¹, Rodrigo CARBALLAL², Carmen MONTERROSO³, Julia SÁNCHEZVILAS⁴, Miguel SERRANO² and Santiago ORTIZ²</p> <p>1. Department of Ecology and Animal Biology, University of Vigo, Spain 2. Department of Botany, University of Santiago de Compostela, Spain 3. Department of Soil Science and Agricultural Chemistry, University of Santiago de Compostela, Spain 4. Organisms and Environment Division, Cardiff University, UK</p>

S.4	Friday, June 16th (13:40– 16:00) Chairs: Dr Sandrine ISNARD & Prof Stefan SIEBERT	
Regular talk 1 13:50	15'	Revegetation of mine tailings in the abandoned asbestos mine of Balangero and Corio (Torino, Italy): Twenty years after Sergio FAVERO-LONGO^{1,2}, Gianluca CHIEPPA¹, Laura GUGLIELMONE¹, Antonio GHIONE² <i>1. Department of Life Sciences and Systems Biology, University of Torino, Italy</i> <i>2. Interdepartmental Center 'G. Scansetti' for Studies on Asbestos and Other Toxic Particulates, University of Torino, Italy</i> <i>3. R.S.A. S.r.l., Balangero, Italy</i>
Regular talk 2 14:05	15'	Characterising soil microbial diversity for conservation and restoration using large-scale DNA-based methods in New Caledonian ultramafic ecosystems Fabian CARRICONDE¹, Laurent MAGGIA^{2,3}, Alexia STOKES⁴, Monique GARDES⁵, Kelly DINH¹, Nicolas FERNANDEZ NUNEZ^{1,6}, Julien DEMENOIS^{1,7,8,9}, Pierre-Louis STENGER¹, Julie RIPOLL^{1,10}, Audrey LEOPOLD¹, Jenifer READ¹¹ and Pierre MOURNET^{2,3} <i>1. Equipe SolVeg, Institut Agronomique néo-Calédonien (IAC), Nouvelle-Calédonie</i> <i>2. CIRAD, UMR AGAP Institut, France</i> <i>3. CIRAD, UMR AGAP Institut, INRAE, Institut Agro, Université de Montpellier, France</i> <i>4. UMR AMAP, INRAE, Université de Montpellier, IRD, CIRAD, CNRS, France</i> <i>5. UMR EDB, Université Toulouse III – Paul Sabatier, CNRS, IRD, France</i> <i>6. UMI TransVIHMI, Université de Montpellier, INSERM, IRD, France</i> <i>7. AIDA, Université de Montpellier, CIRAD, France</i> <i>8. CIRAD, UPR AIDA, Costa Rica</i> <i>9. CATIE, Centro Agronómico Tropical de Investigación y Enseñanza, Costa Rica</i> <i>10. LIRMM, Université de Montpellier, CNRS, France</i> <i>11. School of Biological Sciences, Monash University, Australia</i>
Regular talk 3 Tuesday, June 13th 15:00	15'	Even edaphic specialists can die from climate extremes: Plant mortality on ultramafic soils after a heatwave and intense drought Lorenzo LAZZARO¹, Andrea COPPI¹, Federico SELVI² <i>1. Department of Biology, University of Florence, Italy</i> <i>2. Department of Agriculture, Food, Environment and Forestry, Laboratories of Botany, University of Florence, Italy</i>
Regular talk 4 15:05	15'	Implementing Access and Benefit Sharing (ABS) of Genetic Resources (GR) for Agromining in Sabah, Malaysia Gerald JETONY¹, Guillaume ECHEVARRIA² and Julenah AG. NUDDIN³ <i>1. Sabah Biodiversity Centre</i> <i>2. Laboratoire Sols et Environnement, INRAE, Université de Lorraine</i> <i>3. Universiti Teknologi MARA</i>
Regular talk 5 15:20	15'	Plant communities of Cuban ultramafic soils and the impacts of climate change Rosalina BERAZAÍN ITURRALDE¹ and Ilsa María FUENTES MARRERO² <i>1. Jardín Botánico Nacional, University of Habana, Cuba</i> <i>2. Institute of Ecology and Sistematic, CITMA, Cuba</i>

Regular talk 6 15:35	15'	Reproductive life traits of plants growing on ultramafic substrate on the Koniambo massif (New Caledonia) as a tool to reinforce and reconnect fragmented forests Bruno FOGLIANI¹, Yawiya ITITIATY², Aurore MARTINI² and Fabrice BRESCIA² <i>1. ISEA, University of New Caledonia, New Caledonia</i> <i>2. ARBOREAL, New Caledonian Agronomic Institute, New Caledonia</i>
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S5

ORAL SESSION

SESSION 5:

Ecophysiology, molecular biology, and genetics

S.5	Thrusday, June 15 th (14:00 – 17:00) Chairs: Prof Joe POLLARD & Prof Mark AARTS	
Regular talk 1 14:00	15'	Addressing the diversity of the mechanisms involved in nickel hyperaccumulation Sylvain MERLOT^{1,5}, Dubiel ALFONSO GONZALEZ^{2,3}, Rolando REYES FERNANDEZ³, Celestine BELLOEIL¹, Vanesa GARCIA DE LA TORRE¹, Sébastien THOMINE¹ and Yohan PILLON⁴ <i>1. Institute for Integrative Biology of the Cell, CEA, CNRS, Paris- Saclay University, France</i> <i>2. Jardín Botánico Nacional, Universidad de La Habana, Cuba</i> <i>3. Agronomy Faculty, Agrarian University of La Havana, Cuba</i> <i>4. Laboratory of Tropical and Mediterranean Symbiosis, IRD, CIRAD, INRA, Montpellier University, France</i> <i>5. Laboratoire de Recherche en Sciences Végétales, Toulouse University Paul Sabatier, CNRS, INP Toulouse, France</i>
Regular talk 2 14:15	15'	Development of genetic tools for analysis of Ni/Zn/Cd hyperaccumulation in <i>Noccaea caerulea</i> Jitpanu YAMJABOK, Laurens VAN OOSTROM, Lies VAN DER HEIJDEN, Hedayat BAGHERI, Antony VAN DER ENT, Joost VAN DEN HEUVEL, Mark G.M. AARTS <i>Laboratory of Genetics, Wageningen University & Research, The Netherlands</i>
Regular talk 3 14:30	15'	Does polyploidy affect responses to Nickel in Ni-accumulating plants? Insights from the model species <i>Odontarrhena bertolonii</i> (Brassicaceae) Ilaria COLZI¹, Cristina GONNELLI¹ and Federico SELVI² <i>1. Department of Biology, University of Florence, Italy</i> <i>2. Department of Agriculture, Food, Environment and Forestry, University of Florence, Italy</i>
Regular talk 4 14:45	15'	Serpentine Tolerance in <i>Mimulus guttatus</i>: Gradual Acclimation to Low Ca:Mg Emily PALM¹, and Elizabeth VAN VOLKENBURGH² <i>1. Department of Biotechnology and Biosciences, University of Milan-Bicocca, Italy</i> <i>2. Department of Biology, University of Washington, USA</i>

Regular talk 5 15:00	15'	<p>The Role of Free L-Histidine in Ni and Zn Translocation in Hyperaccumulator and Non-accumulator Brassicaceae Henk SCHAT¹, Anna KOZHEVNIKOVA² and Ilya SEREGIN²</p> <p>1. Laboratory of Genetics, Wageningen University, The Netherlands 2. Timryazev Institute of Plant Physiology, RAS Moscow, Russia</p>
Regular talk 6 15:45	15'	<p>X-ray Fluorescence herbarium ionomics on plants from ultramafic, calcareous, and volcanic soils in Japan Takafumi MIZUNO¹, Daichi KONDO², Hiroto KASAI², Imam PURWADI³, Antony VAN DER ENT^{3,4}, Yoshinori MURAI⁵, Atsushi HASHIMOTO¹ and Toshihiro WATANABE⁶</p> <p>1. Graduate School of Bioresources, Mie University, Japan 2. Faculty of Bioresources, Mie University, Japan 3. Sustainable Minerals Institute, The University of Queensland, Australia 4. Laboratory of Genetics, Wageningen University & Research, The Netherlands 5. Department of Botany, National Museum of Nature and Science, Japan 6. Research Faculty of Agriculture, Hokkaido University, Japan</p>
Regular talk 7 16:00	15'	<p>Indium Knocks at the Door: The Mechanism of Rice Inhibition by Emerging Contamination Elements Boon Huat CHEAH¹, Pei-Chu LIAO¹, Jing-Chi LO², Yu-Tsen WANG, I-Chien TANG, Kuo-Chen YEH, Dar-Yuan LEE and Ya-Fen LIN</p> <p>1. Department of Agronomy, National Taiwan University, Taiwan 2. Department of Horticulture and Biotechnology, Chinese Culture University, Taiwan 3. Agricultural Biotechnology Research Center, Academia Sinica, Taiwan 4. Department of Agricultural Chemistry, National Taiwan University, Taiwan</p>
Regular talk 8 16:15	15'	<p>The hyperaccumulator <i>Leucocroton havanensis</i> overexpress two Ferroportins/IREGs involved in nickel transportation Dubiel ALFONSO GONZÁLEZ^{1,3}, Vanesa GARCÍA DE LA TORRE², Rolando REYES FERNÁNDEZ³, Sébastien THOMINE² and Sylvain MERLOT²</p> <p>1. Jardín Botánico Nacional, University of Habana, Cuba 2. Institute for Integrative Biology of the Cell, CEA, CNRS, Paris- Saclay University, France 3. Agronomy Faculty, Agrarian University of La Havana, Cuba</p>
Regular talk 9 16:30	15'	<p>Leaf elemental composition of species growing on contrasting soils in two adjacent rainforest: serpentinized ultramafic versus volcano-sedimentary rock Tanguy JAFFRE F^{1,2}, Sandrine ISNARD² and Thomas IBANEZ²</p> <p>1. UMR AMAP, IRD, Herbar de Nouvelle-Calédonie, Nouméa, New Caledonia 2. UMR AMAP, Univ. Montpellier, CNRS, IRD, CIRAD, INRAE, Montpellier, France</p>
Regular talk 10 16:45	15'	<p>Physiological response in relation to nickel hyperaccumulation in <i>Bornmuellera emarginata</i> cultivated in hydroponics over a nickel gradient Serigne LY¹, Guillaume ECHEVARRIA^{1,2}, Mark G.M. AARTS³, Stéphanie OUVRARD¹ and Antony VAN DER ENT^{1,2,3}</p> <p>1. Laboratoire Sols et Environnement, Université de Lorraine-INRAE, France 2. Centre for Mined Land Rehabilitation, Sustainable Minerals Institute, The University of Queensland, Queensland, Australia. 3. Laboratory of Genetics, Wageningen University and Research, The Netherlands</p>

S.6	Thursday, June 15 th (08:30 – 11:40) Chairman: Prof Roger REEVES & Prof Alan BAKER	
Keynote 08:45	40'	The Discovery and Global Distribution of Hyperaccumulator Plants Roger REEVES <i>Palmerston North, New Zealand</i>
Regular talk 1 09:25	15'	Exploring the associations between soil microbiome, root architecture, and Zn & Cd accumulation in <i>Arabidopsis halleri</i>: results from a transplant experiment in rhizoboxes Alicja BABST-KOSTECKA¹, Kamila MURAWSKA-WLODARCZYK¹, Priyanka KUSHWAHA¹, Yongjian CHEN¹, Charlotte DIETRICH², and Manzeal KHANAL¹ <i>1. Department of Environmental Science, The University of Arizona, United States</i> <i>2. W. Szafer Institute of Botany, Polish Academy of Science, Poland</i>
Regular talk 2 09:40	15'	Metal Hyperaccumulators of Mexico and Central America: A Survey of Recent Progress Joe POLLARD¹, Dulce NAVARRETE GUTIÉRREZ^{2,3}, Guillaume ECHEVARRIA^{3,4}, Grace MCCARTHA^{1,5}, Haley DISINGER¹ and Alan BAKER⁶ <i>1. Department of Biology, Furman University, USA</i> <i>2. Departamento de Fitotecnia, Universidad Autonoma Chapingo, Mexico</i> <i>3. Laboratoire Sols et Environnement, Université de Lorraine, France</i> <i>4. ECONICK, France</i> <i>5. Natural Heritage New Mexico, The University of New Mexico, USA</i> <i>6. School of BioSciences, The University of Melbourne, Australia</i>
Regular talk 3 09:55	15'	Trace element hyperaccumulator plant traits: a call for trait data collection Béatrice GERVAIS-BERGERON¹, Adrian PAUL¹, Pierre-Luc CHAGNON², Alan BAKER^{3,4,5}, Antony VAN DER ENT^{3,5}, Michel-Pierre FAUCON⁶, Celestino QUINTELA-SABARÍS⁷ and Michel LABRECQUE¹ <i>1. Institut de recherche en biologie végétale, Université de Montréal, Canada</i> <i>2. Agriculture and Agrifood Canada, Canada</i> <i>3. Sustainable Minerals Institute, The University of Queensland, Australia</i> <i>4. School of BioSciences, The University of Melbourne, Australia</i> <i>5. Laboratoire Sols et Environnement, Université de Lorraine, France</i> <i>6. UniLaSalle, AGHYLE, France</i> <i>7. Grupo de Ecología Animal (GEA), Universidad de Vigo, Spain</i>
Regular talk 4 10:10	15'	XRF ionomics of herbarium specimens from the Balkan Peninsula Tomica MIŠLJENOVIC¹, Ksenija JAKOVljeVIC¹, Gordana TOMOVIĆ¹, Alan BAKER^{2,3,4,6}, Antony VAN DER ENT^{2,3,5,6} and Guillaume ECHEVARRIA^{2,3,6} <i>1. Institute of Botany and Botanical Garden, Faculty of Biology, University of Belgrade, Serbia</i> <i>2. Laboratoire Sols et Environnement, INRAE, Université de Lorraine, France</i> <i>3. Centre for Mined Land Rehabilitation, Sustainable Minerals Institute, The University of Queensland, Queensland, Australia</i> <i>4. School of BioSciences, The University of Melbourne, Parkville, VIC 3010, Australia</i> <i>5. Laboratory of Genetics, Wageningen University and Research, The Netherlands</i> <i>6. Econick, Nancy, France</i>

<p>Regular talk 5 11:00</p>	<p>15'</p>	<p>Nickel accumulation in plants from Shebenik mountain massif Aida BANI¹, Ermelinda GJETA², Vjollca IBRO³, Dolja PAVLOVA⁴, SeitSHALLARI¹, Federico SELVI⁵, Christina HIPFINGER⁶, Marku PUSCHENREITER⁶, Guillaume ECHEVARRIA⁷ 1. <i>Department of Environment and Natural Resources. Faculty of Agronomy and Environment, Agricultural University of Tirana, Tirane, Albania</i> 2. <i>Department of Biology, Aleksander Xhuvani University, Elbasan</i> 3. <i>Department of Crop Production, Faculty of Agronomy and Environment, Agricultural University of Tirana, Tirane, Albania</i> 4. <i>Department of Botany, Faculty of Biology, University of Sofia, Sofia 1504, Bulgaria</i> 5. <i>Department of Agrifood Production and Environmental Sciences, University of Florence, P.le delle Cascine 28, 50144 Firenze, Italy</i> 6. <i>Department of Forest and Soil Sciences, Institute of Soil Research, University of Natural Resources and Life Sciences, Vienna, Austria</i> 7. <i>Laboratoire Sols et Environnement, Université de Lorraine, INRAE, 54000 Nancy, France</i></p>
<p>Regular talk 6 11:15</p>	<p>15'</p>	<p>Biogeochemistry of the Serpentine of Rhodes and Cyprus Roger REEVES¹, Maria ALOUPI², Emmanouil DAFTSIS³, John STRATIS³, Petros MASTORAS² and Panayiotis DIMITRAKOPOULOS² 1. <i>Palmerston North, New Zealand</i> 2. <i>Department of Environment, University of the Aegean, Mytilene, Greece</i> 3. <i>Laboratory of Analytical Chemistry, Aristotle University of Thessaloniki, Greece</i></p>

S.7	Tuesday, June 13th (13:50 – 15:30) Chairs: Prof Nishanta RAJAKARUNA & Dr Antony VAN DER ENT	
Keynote 13:35	40'	Lessons on Ecology and Evolution from the Study of Edaphic Specialization Nishanta RAJAKARUNA^{1,2} 1. <i>Biological Sciences Department, California Polytechnic State University, USA</i> 2. <i>Unit for Environmental Sciences and Management, North-West University, South Africa</i>
Regular talk 1 14:15	15'	Current update on floristic diversity of Malagasy inselbergs Marina RABARIMANARIVO¹, Sylvie ANDRIAMBOLOLONERA¹ and Stefan POREMBSKI² 1. <i>Research Unit, Missouri Botanical Garden Madagascar Program, Madagascar</i> 2. <i>Department of Botany and Botanical Garden, Rostock University, Germany</i>
Regular talk 2 14:30	15'	Genomic basis of tolerance to extremely low-pH in an extremophyte (<i>Carex angustisquama</i>, Cyperaceae) in highly acidic volcanic soils in Japan Koki NAGASAWA¹, Hiroaki SETOGUCHI¹, Ken NAITO², Atsushi NAGANO^{3,4}, Naoko ISHIKAWA⁵ and Shota SAKAGUCHI¹ 1. <i>Graduate School of Human and Environmental Studies, Kyoto University, Japan</i> 2. <i>Research Center of Genetic Resources, National Agriculture and Food Research Organization, Japan</i> 3. <i>Faculty of Agriculture, Ryukoku University, Japan</i> 4. <i>Institute for Advanced Biosciences, Keio University, Japan</i> 5. <i>Graduate School of Science, Osaka Metropolitan University, Japan</i>
Regular talk 3 14:45	15'	Setting an agenda for gypsum ecosystem research in southern Africa Stefan SIEBERT¹, Sara PALACIO^{1,2}, Arantzazu LUZURIAGA³, Gillian MAGGS-KÖLLING^{1,4}, Eugene MARAIS⁴, Silvia MATESANZ³, María PRIETO³, Yolanda PUEYO⁵, Nishanta RAJAKARUNA^{1,6}, Ana SÁNCHEZ³, Sarina CLAASSENS¹ 1. <i>Unit for Environmental Sciences and Management, North-West University, Potchefstroom, South Africa</i> 2. <i>Instituto Pirenaico de Ecología (IPE-CSIC), Jaca, Huesca, Spain</i> 3. <i>Área Biodiversidad y Conservación, Departamento de Biología, Geología, Física Aplicada y Química Inorgánica, ESCET, Universidad Rey Juan Carlos, Móstoles, Madrid, Spain</i> 4. <i>Gobabeb - Namib Research Institute, Namibia</i> 5. <i>Instituto Pirenaico de Ecología (IPE, CSIC), Zaragoza, Spain</i> 6. <i>Biological Sciences Department, California Polytechnic State University, San Luis Obispo, United States</i>

S.8	Friday, June 16th (14:55 – 15:20) Chairman: Prof Alan BAKER, Dr Rufus CHANEY & Prof Jean-Louis MOREL	
Keynote 08:30	40'	Many Reasons for Study of Serpentine Ecology Rufus L. CHANEY <i>Chaney Environmental LLC, USA</i>
Regular talk 1 09:15	15'	Agromining in Sabah, Malaysia: Current Status of Knowledge and Future Potentials Julenah AG. NUDDIN¹, Rimi REPIN², Antony VAN DER ENT³, Guillaume ECHEVARRIA⁴ <i>1. Faculty of Applied Sciences, Universiti Teknologi MARA Cawangan Sabah, Kota Kinabalu, Sabah, Malaysia</i> <i>2. Sabah Parks, Kota Kinabalu, Sabah, Malaysia</i> <i>3. Department of Plant Sciences, Wageningen University & Research, Wageningen, Netherlands</i> <i>4. Soils and Environment Laboratory, University of Lorraine, Vandoeuvre-lès-Nancy, France</i>
Regular talk 2 09:30	15'	Developing speed-breeding techniques for European nickel metal crops Mirko SALINITRO¹, Antony van der ENT^{2,3,4}, Guillaume ECHEVARRIA^{3,4}, Annalisa TASSONI¹ <i>1. Department of Biological, Geological and Environmental Sciences, University of Bologna, Italy</i> <i>2. Laboratory of Genetics, Wageningen University and Research, The Netherlands.</i> <i>3. Centre for Mined Land Rehabilitation, Sustainable Minerals Institute, The University of Queensland, Queensland, Australia.</i> <i>4. Laboratoire Sols et Environnement, INRAE, Université de Lorraine, France</i>
Regular talk 3 09:45	15'	Environmental assessment tools applied to the agromining chain, from the field to marketable products Marie-Odile SIMONNOT¹, Baptiste LAUBIE¹, Bastien JALLY and Marie-Noëlle PONS³ <i>1. Université de Lorraine, CNRS, LRGP, 54000 Nancy, France</i>
Regular talk 4 10:00	15'	Nickel accumulation and spatial distribution of metals assessed by SR- μXRF in tropical hyperaccumulators Luiz LIMA¹, Sarah NICHOLAS², Ryan TAPPERO², Guillaume ECHEVARRIA³, Clístenes NASCIMENTO¹ <i>1. Department of Agronomy, Federal Rural University of Pernambuco, Brazil</i> <i>2. National Synchrotron Light Source II, Brookhaven National Laboratory, United States</i> <i>3. INRAE, Soil & Environment Laboratory, France</i>

Regular talk 5 10:45	15'	<p>Optimal co-cropping densities of <i>Noccaea caerulea</i> with <i>Sedum plumbizincicola</i> for zinc <i>in situ</i> phytoextraction depend on metal bioavailability</p> <p>Julien JACQUET^{1,2}, Gabrielle MICHAUDEL¹, Emile BENIZRI², Guillaume ECHEVARRIA^{1,2,3} and Catherine SIRGUEY²</p> <p>1. Econick, France 2. Laboratoire Sols et Environnement, Université de Lorraine, INRAE, France 3. Centre for Mined Land Rehabilitation, Sustainable Minerals Institute, University of Queensland, Australia</p>
Regular talk 6 11:00	15'	<p>Biotechnological tools for improving Ni agromining in ultramafic soils</p> <p>Ángeles PRIETO-FERNÁNDEZ¹, Tania PARDO^{1,2}, Beatriz RODRÍGUEZ-GARRIDO¹, Emile BENIZRI³, and Petra KIDD¹</p> <p>1. Misión Biológica de Galicia (MBG) Sede Santiago de Compostela, CSIC, Spain 2. Unit of Plant Protection Products, INIA-CSIC, Spain 3. Laboratoire Sols et Environnement, Université de Lorraine, INRAE, France</p>
Regular talk 7 11:15	15'	<p>Improving plant growth on mine tailings by using N-rich residual materials from mine water treatment</p> <p>Ricot ST-AIMÉ, Marie GUITTONNY and Carmen Mihaela NECULITA</p> <p>Research Institute on Mines and the Environment, Université du Québec en Abitibi-Témiscamingue, Canada</p>
Regular talk 8 11:30	15'	<p>LIFE-AGROMINE, a pilot-scale demonstration carried out at EU level</p> <p>Guillaume ECHEVARRIA^{1,2,3}, Aida BANI^{4,5}, Claire HAZOTTE³, Petra KIDD⁶, Maria KONSTANTINO⁷, Tom KUPPENS⁸, Gaylord MACHINET⁹, Jean Louis MOREL¹, Ángeles PRIETO-FERNÁNDEZ⁶, Markus PUSCHENREITER¹⁰, Marie-Odile SIMONNOT¹¹, Evgenia TSIANOU¹²</p> <p>1. Université de Lorraine, INRAE, Laboratoire Sols et Environnement, France 2. University of Queensland, Sustainable Minerals Institute, Centre for Mined Land Rehabilitation, Australia 3. ECONICK, France 4. Agro-Environment and Economic Management Center, Albania 5. Agricultural University of Tirana, Albania 6. CSIC, Instituto de Investigaciones Agrobiológicas de Galicia, Spain 7. Department of Agriculture, International Hellenic University, Greece 8. University Hasselt, Belgium 9. MICROHUMUS, France 10. BOKU, Department of Forest and Soil Sciences, Institute of Soil Research, Austria 11. Université de Lorraine, CNRS, Laboratoire Réactions et Génie des Procédés, France 12. Alchemia Nova, Austria</p>
Regular talk 9 11:45	15'	<p>Preliminary results of optimizing Ni phytoextraction by <i>Odontarrhena lesbiaca</i></p> <p>Ioanna CHATZELLI, Maria ALOUPI and Panayiotis G. DIMITRAKOPOULOS</p> <p>Department of Environment, University of the Aegean, Greece</p>

Regular talk 10 12:00	15'	Changes on soil bacterial communities after an agromining field trial in the NW of the Iberian Peninsula Vanessa ÁLVAREZ-LÓPEZ¹, Ángeles PRIETO-FERNÁNDEZ², Beatriz RODRÍGUEZ-GARRIDO², Carmela MONTERROSO³, Guillaume ECHEVARRIA⁴ and Petra KIDD^{2†} <i>1. AQUATERRA - Gestión sostenible de los recursos hídricos y del suelo. CICA - Centro Interdisciplinar de Química e Biología. Facultad de Ciencias University of A Coruña, Spain</i> <i>2. Suelos Biosistemas y Ecología Agroforestal, MBG-CSIC sede Santiago de Compostela, Spain</i> <i>3. Departamento de Edafología e Química Agrícola, University of Santiago de Compostela, Spain</i> <i>4. Soils and Environment Laboratory, University of Lorraine, France</i> <i>†. Deceased</i>
Regular talk 11 13:15	15'	Substrate formulation for agromining of cobalt from aged mine tailings Samuel TEILLAUD^{1,2}, Lucie COUDERT¹, Marie GUITTONNY¹, Jean Louis MOREL^{1,3}, Baptiste LAUBIE² and Marie-Odile SIMONNOT² <i>1. Institut de Recherche en Mines et en Environnement, Université du Québec en Abitibi-Témiscamingue, Canada</i> <i>2. Laboratoire Réaction et Génie des Procédés, CNRS, Université de Lorraine, France</i> <i>3. Laboratoire Sols et Environnement, INRAE, Université de Lorraine, France</i>
Regular talk 12 13:30	15'	Assisting nickel agromining using sustainable amendments Alexis DURAND, Léa JAFEU, Pierre LEGLIZE and Emile BENIZRI <i>Laboratoire Sols et Environnement, Université de Lorraine, INRAE, France</i>

S1

POSTER EXHIBITION

SESSION 1:

Geology, soils & biogeochemical cycles

Session 1 - posters

S1.P1	Application of clay minerals in remediation of heavy metal pollution in soil Teuta BUSHI¹, Aida BANI <i>1. Department of Physics, Faculty of Forest Sciences Agricultural University of Tirana, Albania</i> <i>2. Department of Agro-Environment and Ecology, Faculty of Agronomy and Environment, Agricultural University of Tirana, Albania</i>
S1.P2	Assessing ecological, environmental, and human health risks associated with ultramafic soils in Brazil Luiz LIMA¹, Clístenes NASCIMENTO² <i>1. Department of Agronomy, Federal Rural University of Pernambuco, Brazil</i>
S1.P3	Distribution of Heavy Metals in the Different Compartments in Lake Dushku, Albania Aida BANI¹, Dolja PAVLOVA², Seit SHALLARI¹ <i>1. Department of Environment and Natural Resources. Faculty of Agronomy and Environment, Agricultural University of Tirana, Tirana, Albania</i> <i>2. Department of Botany, Faculty of Biology, University of Sofia, Sofia 1504, Bulgaria</i>

S1.P4	<p>Interactions of Cr(III)/Cr(VI) Mixtures with Freshwater Algae: a Stable Isotope Approach Elsa SALLES¹, Armand MAUL¹, Carole COSSU-LEGUILLE, Thi Tuyen NGUYEN², Maximilien BEURET¹, Alexis GROLEAU², Yann SIVRY², Yulia ERBAN KOCHERGINA³, Pavel KRAM³, Vincent NORMANT¹ and Davide VIGNATI¹</p> <p>1. LIEC, Université de Lorraine, CNRS, France 2. IPGP, Université Paris Cité, CNRS, France 3. Division of Geochemistry and Laboratories, Czech Geological Survey, Czech Republic</p>
S1.P5	<p>The influence of fire and fire retardant (Phos-chek®) on plant diversity and non-native species abundance in California's serpentine chaparral Zach RAPOSO^{1,2}, Chris HOWINGTON³, Alex PENA⁴, Luka NEGOITA⁵, Anthony FERRERO⁶, Peter WALSH⁶, Dylan STEPHENS⁶ and Nishanta RAJAKARUNA^{6,7}</p> <p>1. Geospatial & Environmental Department, Althouse and Meade, Inc., USA 2. Natural Resources Management and Environmental Sciences, California Polytechnic State University, USA 3. Natural Resources Conservation Service, United States Department of Agriculture, USA 4. United States Forest Service, United States Department of Agriculture, USA 5. Charles Darwin Research Station, Charles Darwin Foundation, Ecuador 6. Biological Sciences Department, California Polytechnic State University San Luis Obispo, USA 7. Unit for Environmental Sciences and Management, North-West University, South Africa</p>
S1.P6	<p>Wettability of serpentine and other minerals upon rising temperatures Sara NEGRI¹, Mohammad Ali MONAJJEM^{1,2}, Marta IANNICELLI¹ and Eleonora BONIFACIO¹</p> <p>1. Department of Agricultural, Forest and Food Sciences, University of Torino, Italy 2. Soil Science Department, University of Tehran, Karaj, Iran</p>
S1.P7	<p>Geochemical classification of <i>Macrotermes falciger</i> termite mounds on laterite profile Serge Ilunga Ngoy¹, Yannick Callec², Denis Thieublemont², Apolline Auclerc¹, Françoise Watteau¹</p> <p>1. Laboratoire Sols et Environnement (LSE-INRAE), INRAE/Université de Lorraine (UL), France 2. Bureau de Recherches Géologiques et Minières (BRGM), France</p>

S2

POSTER EXHIBITION

SESSION 2:

Biodiversity: taxonomy and systematics

Session 2 - posters

S2.P1	<p>Bacterial and fungal diversity found in serpentine and non-serpentine soil crusts from two different climatic regions in South Africa Danielle BOTHA¹, Arthurita VENTER¹, Sandra BARNARD¹, Sarina CLAASSENS¹, Nishanta RAJAKARUNA^{1,2} and Stefan SIEBERT¹</p> <p>1. Unit for Environmental Sciences and Management, North-West University, South Africa 2. Biological Sciences Department, California Polytechnic State University, USA</p>
S2.P2	<p>New <i>Stylochaeton</i> species from ultramafic soil in South Africa Madeleen STRUWIG¹, Stefan SIEBERT² and Abraham VAN WYK³</p> <p>1. Unit for Environmental Sciences and Management, North-West University, Mafikeng Campus, South Africa 2. Unit for Environmental Sciences and Management, North-West University, South Africa. 3. H.G.W.J. Schweickerdt Herbarium, Department of Plant and Soil Sciences, University of Pretoria, South Africa</p>

S2.P2	<p>New <i>Stylochaeton</i> species from ultramafic soil in South Africa Madeleen STRUWIG¹, Stefan SIEBERT² and Abraham VAN WYK³</p> <p>1. <i>Unit for Environmental Sciences and Management, North-West University, Mafikeng Campus, South Africa</i> 2. <i>Unit for Environmental Sciences and Management, North-West University, South Africa.</i> 3. <i>H.G.W.J. Schweickerdt Herbarium, Department of Plant and Soil Sciences, University of Pretoria, South Africa</i></p>
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S3

POSTER EXHIBITION

SESSION 3:

Ecology and evolution

Session 3 - posters

S3.P1	<p>Lichen diversity and abundance on serpentinite and granite outcrops of the Slavkov Forest (Czech Republic) Sergio FAVERO-LONGO¹, Elisabetta BIANCHI², Stefano LOPPI², Luca PAOLI³, Zuzana FAČKOVCOVÁ⁴ and Pavel KRÁM⁵</p> <p>1. <i>Department of Life Sciences and Systems Biology, University of Torino, Italy</i> 2. <i>Department of Life Sciences, University of Siena, Italy</i> 3. <i>Department of Biology, University of Pisa, Italy</i> 4. <i>Plant Science and Biodiversity Centre, Slovak Academy of Sciences, Slovakia</i> 5. <i>Global Change Research Institute, Czech Academy of Sciences, Czech Republic</i></p>
S3.P2	<p>Community assembly of serpentine plants is driven by interspecific differences in functional traits Guillaume DELHAYE¹, Panayiotis G. DIMITRAKOPOULOS² and George C. ADAMIDIS³</p> <p>1. <i>Ecosystem Stewardship, Royal Botanic Gardens Kew, United Kingdom</i> 2. <i>Department of Environment, University of the Aegean, Greece</i> 3. <i>Biology Department, University of Patras, Greece</i></p>
S3.P3	<p>Disentangling plant ionomes of two pseudo-metallophyte orchid species on contrasting soils in the Balkans Ksenija JAKOVljević¹, Tomica MIŠLJENović¹, Antony VAN DER ENT^{2,3,4,5}, Jasmina ŠINŽARSEKULIĆ¹, Jelena MUTIĆ⁶, Marija ČOŠIĆ¹, Gordana ANDREJIĆ⁷, Dragana RANĐELOVIĆ⁸ and Vladan DJORDJEVIĆ¹</p> <p>1. <i>Institute of Botany and Botanical Garden, Faculty of Biology, University of Belgrade, Serbia</i> 2. <i>Laboratoire Sols et Environnement, INRAE, Université de Lorraine, France</i> 3. <i>Centre for Mined Land Rehabilitation, Sustainable Minerals Institute, The University of Queensland, Queensland, Australia</i> 4. <i>Laboratory of Genetics, Wageningen University and Research, The Netherlands</i> 5. <i>ECONICK, 1 Rue Grandville 54000 Nancy, France</i> 6. <i>Faculty of Chemistry, University of Belgrade, Serbia</i> 7. <i>Institute for the Application of Nuclear Energy – INEP, University of Belgrade, Serbia</i> 8. <i>Institute for Technology of Nuclear and Other Minerals Raw Materials, Serbia</i></p>
S3.P4	<p>Does flowering phenology promote functional trait differentiation across the serpentine boundary? Noelia HILDALGO-TRIANA¹, Claudia ARRANZ¹, Nazaret KEEN RUIZ¹, Jaime PEREÑA ORTIZ¹, Pablo ARRUFAT², Andrés V. PEREZ-LATORRE¹ and Peter B. PEARMAN^{2,3,4}</p> <p>1. <i>Department of Botany and Plant Physiology, University of Malaga, Spain</i> 2. <i>Department of Plant Biology and Ecology, University of the Basque Country, Spain</i> 3. <i>IKERBASQUE Basque Foundation for Science, Bilbao, Bizkaia, Spain</i> 4. <i>BC3 Basque Center for Climate Change, Leioa, Bizkaia, Spain</i></p>

S3.P6	<p>The effect of substrate type and species richness on biomass production and nickel concentration</p> <p>Panayiotis G. DIMITRAKOPOULOS, Maria ALOUPI, Dimitris SKOPIANOS, and Ioanna CHATZELLI</p> <p><i>Department of Environment, University of the Aegean, Greece</i></p>
S3.P7	<p>Ultramafic lichens: an ecological investigation of an understudied organism in a well-studied system.</p> <p>Michael MULROY¹, Jason DART^{2,3}, Cameron WILLIAMS⁴, Rikke REESE NÆSBORG⁴, Alan FRYDAY⁵, Scott JOHNSTON⁶, and Nishanta RAJAKARUNA^{1,7}</p> <p><i>1. Biological Sciences Department, California Polytechnic State University, San Luis Obispo, USA</i> <i>2. Althouse and Meade, Inc., USA</i> <i>3. Robert F. Hoover Herbarium, California Polytechnic University, San Luis Obispo</i> <i>4. Conservation Department, Santa Barbara Botanic Garden, USA</i> <i>5. Herbarium, Department of Plant Biology, Michigan State University, USA</i> <i>6. Physics Department, California Polytechnic State University, San Luis Obispo, USA</i> <i>7. Unit for Environmental Sciences and Management, North-West University, South Africa</i></p>
S3.P8	<p>A microscopic description of the actinorhizal nodules of Casuarinaceae trees growing on ultramafic areas in Sabah (Malaysia)</p> <p>Celestino QUINTELA-SABARÍS¹, Chor Wai LO², Hasna BOUBAKRI³, Philippe NORMAND³ and Jorge DOMÍNGUEZ¹</p> <p><i>1. Departamento de Ecología e Biología Animal, Universidade de Vigo, Spain</i> <i>2. Fakulti Sains Gunaan, Universiti Teknologi MARA- Cawangan Sabah, Malaysia</i> <i>3. Laboratoire d'Ecologie Microbienne, Université Lyon 1, France</i></p>

S4

POSTER EXHIBITION

SESSION 4:

Conservation and restoration

Session 4 - posters

S4.P1	<p>Germination and initial growth characteristics of <i>Saussurea modesta</i> (Asteraceae), an endangered serpentine plant in Japan</p> <p>Ryusuke INOUE¹, Akihiro YAMAMOTO², Jun WASAKI¹ and Takayuki NAKATUBO^{2,3}</p> <p><i>1. Graduate School of Integrated Sciences for Life, Hiroshima University, Japan</i> <i>2. Division of Cultivation and Exhibition, Hiroshima Botanical Garden, Japan</i> <i>3. Hiroshima University Museum, Hiroshima University, Japan</i></p>
S4.P2	<p>Modeling Species Distributions of Select Obligate Metallophytes in Response to Climate Change</p> <p>Lauren EBERTH¹, Haley DISINGER¹, Joe POLLARD¹ and John QUINN¹</p> <p><i>Department of Biology, Furman University, USA</i></p>
S4.P3	<p>Phytoremediation and Nurse Potential of Aloe Plants from Ultramafic Areas on Mine Tailings</p> <p>João MARCELO-SILVA, Masego RAMABU and Stefan SIEBERT</p> <p><i>Unit for Environmental Sciences and Management, North-West University, Potchefstroom 2520, South Africa</i></p>

S4.P4	<p>Trace metal exclusion mechanisms enhance invasion of Mediterranean ultramafic habitats by <i>Ailanthus altissima</i> (Simaroubaceae)</p> <p>Andrea COPPI¹, Lorenzo LAZZARO¹, Cristina GONNELLI¹, Ilaria COLZI¹, Michele MUGNAI^{1,2}, Isabella BETTARINI^{1,2} and Federico SELVI³</p> <p>1. Department of Biology, University of Florence, Italy 2. NBFC, National Biodiversity Future Center, Palermo 90133 3Department of Dept of Agriculture, Food, Environment and Forestry (DAGRI), University of Florence, Italy</p>
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S5

POSTER EXHIBITION

SESSION 5:

Ecophysiology, molecular biology, and genetics

Session 5 - posters

S5.P1	<p>Assessing Nickel Tolerance and Accumulation in the Central American Hyperaccumulator <i>Blepharidium guatemalense</i> Using Hydroponic Systems</p> <p>Lauren EBERTH¹, Dulce NAVARRETE GUTIÉRREZ^{2,3} and Joe POLLARD¹</p> <p>1. Department of Biology, Furman University, USA 2. Departamento de Fitotecnia, Universidad Autonoma Chapingo, Mexico 3. Laboratoire Sols et Environnement, Université de Lorraine, France</p>
S5.P2	<p>Exclusion and Accumulation: Metallophytes Employ Both Mechanisms to Survive on Serpentine and Mining Habitats of Sekhukhuneland, South Africa</p> <p>Sutapa ADHIKARI¹, João MARCELO-SILVA¹, Nishanta RAJAKARUNA^{1,2} and Stefan SIEBERT¹</p> <p>1. Unit for Environmental Sciences and Management, North-West University, South Africa 2. Biological Sciences Department, California Polytechnic State University, United States</p>
S5.P3	<p>Ion profiles of plant species distributed in two contrasting soils: serpentine soil and volcanic acid soil, western Japan</p> <p>Akihiro YAMAMOTO¹, Ryusuke INOUE², Toshihiro WATANABE³, Takayuki NAKATSUBO^{2,4} and Jun WASAKI²</p> <p>1. Division of Cultivation and Exhibition, Hiroshima Botanical Garden, Japan 2. Graduate School of Integrated Sciences for Life, Hiroshima University, Japan 3. Research Faculty of Agriculture, Hokkaido University, Japan 4. Hiroshima University Museum, Hiroshima University, Japan</p>

S6

POSTER EXHIBITION

SESSION 6: Metal hyperaccumulation discovery & monitoring

Session 6 - posters

S6.P1	<p>Element Accumulation by the Holoparasitic Species <i>Cuscuta planiflora</i> Ten. from Serpentine in Bulgaria</p> <p>Dolja PAVLOVA¹, Irina KARADJOVA² and Aida BANI³</p> <p>1. Department of Botany, Faculty of Biology, University of Sofia, Bulgaria 2. Department of Analytical chemistry, Faculty of Chemistry and Pharmacy, University of Sofia, Bulgaria 3. Department of Environment and Natural Resources, Faculty of Agronomy and Environment, Agricultural University of Tirana, Albania</p>
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S6.P2	<p>Assessing the metal accumulation capacity of plants to optimize phytoextraction of mine tailings in arid and semi-arid ecosystems</p> <p>Tomasz WLODARCZYK¹, Kamila MURAWSKA-WLODARCZYK¹, Yves-Marie LEGRAND³, Claude GRISON³, Arthur LASBLEIZ³, Charlotte PEARSON², Raina MAIER¹ and Alicja BABST-KOSTECKA¹</p> <p>1. <i>Department of Environmental Science, The University of Arizona, United States</i> 2. <i>Laboratory of Tree-Ring Research, The University of Arizona, United States</i> 3. <i>Laboratoire de Chimie Bio-Inspirée et d'Innovations Ecologiques, CNRS-Université de Montpellier, France</i></p>
S6.P3	<p>Atomic Spectroscopic Analysis of Macronutrients and Trace Elements in Potential Hyperaccumulators from Sabah</p> <p>N. S. SALIM¹, M. MG BUANG², S. SUMAIL², Antony VAN DER ENT³, Julenah AG. NUDDIN¹ and Guillaume Echevarria⁴</p> <p>1. <i>Faculty of Applied Science Universiti Teknologi MARA Kota Kinabalu, Malaysia</i> 2. <i>Herbarium, Sabah Parks, Kota Kinabalu, Malaysia</i> 3. <i>Department of Plant Sciences, Wageningen University & Research, Netherlands</i> 4. <i>Laboratoire Sols et Environnement, University of Lorraine, France</i></p>
S6.P4	<p>Does metal hyperaccumulation influence leaf traits? Morphological and anatomical investigation of the ultramafic flora of New Caledonia</p> <p>Lily CHENG-CLAVEL & Sandrine ISNARD</p> <p><i>UMR AMAP, Univ. Montpellier, CNRS, IRD, CIRAD, INRAE, Montpellier, France</i></p>
S6.P5	<p>Herbarium and Field Studies of Nickel-Hyperaccumulating Plants from Ultramafic Soils in Guatemala</p> <p>Haley DISINGER¹, Dulce NAVARRETE GUTIÉRREZ^{2,3}, Guillaume ECHEVARRIA^{3,4}, Alan BAKER⁵ and Joe POLLARD¹</p> <p>1. <i>Department of Biology, Furman University, USA</i> 2. <i>Departamento de Fitotecnia, Universidad Autonoma Chapingo, Mexico</i> 3. <i>Laboratoire Sols et Environnement, Université de Lorraine, France</i> 4. <i>ECONICK, France</i> 5. <i>School of BioSciences, The University of Melbourne, Australia</i></p>
S6.P6	<p>Hyperaccumulation of Ni and Zn in <i>Noccaea</i> herbarium specimens</p> <p>Ksenija JAKOVljević¹, Tomica MIŠljENović¹, Alan BAKER^{2,3,4,5}, Antony VAN DER ENT^{2,3,5,6}, Vanessa INVERNON⁷ and Guillaume ECHEVARRIA^{2,3,5}</p> <p>1. <i>Institute of Botany and Botanical Garden, Faculty of Biology, University of Belgrade, Serbia</i> 2. <i>Laboratoire Sols et Environnement, INRAE, Université de Lorraine, France</i> 3. <i>Centre for Mined Land Rehabilitation, Sustainable Minerals Institute, The University of Queensland, Queensland, Australia</i> 4. <i>School of BioSciences, The University of Melbourne, VIC 2010, Australia</i> 5. <i>Econick, Nancy, France</i> 6. <i>Laboratory of Genetics, Wageningen University and Research, The Netherlands</i> 7. <i>National Museum of Natural History, France</i></p>
S6.P7	<p>Ni hyperaccumulation in the flora of Barro Alto, Goiás State, Brazil</p> <p>Leide ANDRADE¹, Fabiana AQUINO¹, Zenilton MIRANDA¹, Cícero PEREIRA¹, Juaci MALAQUIAS¹, and Guillaume ECHEVARRIA^{2,3}</p> <p>1. <i>Embrapa Cerrados, Brazil</i> 2. <i>INRAE, Laboratoire Sols et Environnement, France</i> 3. <i>University of Queensland, Sustainable Minerals Institute, Centre for Mined Land Rehabilitation, Australia</i></p>

S6.P8	<p>Biotechnological tools for improving Ni agromining in ultramafic soils Ángeles PRIETO-FERNÁNDEZ¹, Tania PARDO^{1,2}, Beatriz RODRÍGUEZ-GARRIDO¹, Emile BENIZRI³, and Petra KIDD¹</p> <p>1. <i>Misión Biológica de Galicia (MBG) Sede Santiago de Compostela, CSIC, Spain</i> 2. <i>Unit of Plant Protection Products, INIA-CSIC, Spain</i> 3. <i>Laboratoire Sols et Environnement, Université de Lorraine, INRAE, France</i></p>
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S7

POSTER EXHIBITION

SESSION 7:

Plants of other edaphically challenging

Session 7 - posters

S7.P1	<p>Clay Endemism and Affinity in the California Floristic Province Emma FRYER¹, Alyssa SHON¹, Hannah HUNTLEY¹, Ryan O'DELL² and Nishanta RAJAKARUNA^{1,3}</p> <p>1. <i>Department of Biological Sciences, California Polytechnic State University, USA</i> 2. <i>Bureau of Land Management, United States Department of the Interior, USA</i> 3. <i>Unit for Environmental Sciences and Management, North-West University, South Africa</i></p>
S7.P2	<p>Hyperaccumulator <i>Stanleya pinnata</i>: In Situ Fitness in Relation to Tissue Selenium Concentration Michela SCHIAVON¹, McKenna CASTLEBERRY², Amy WANGELINE³, Bernadette AGUIRRE³, Stefano DALL'ACQUA⁴, Elizabeth PILON-SMITS², and Leonardo LIMA^{2,5}</p> <p>1. <i>Department of Agricultural, Forest and Food Sciences, University of Torino, Italy</i> 2. <i>Biology Department, Colorado State University, USA</i> 3. <i>Biology Department, Laramie County Community College, USA</i> 4. <i>Department of Pharmaceutical and Pharmacological Sciences, University of Padova, Italy</i> 5. <i>Donald Danforth plant Science Center, USA</i></p>
S7.P3	<p>Lessons Learned and Paths Forward: Ensuring Long-Term Success of Revegetation Efforts on Metal-Contaminated Mine Tailings in Arid and Semi-Arid Ecosystems Kamila MURAWSKA-WLODARCZYK¹, Priyanka KUSHWAHA¹, Julian SCHROEDER², Raina MAIER¹ and Alicja BABST-KOSTECKA¹</p> <p>1. <i>Department of Environmental Science, The University of Arizona, United States</i> 2. <i>Department of Cell and Developmental Biology, University of California San Diego, United States</i></p>
S7.P4	<p>The Vertic Clay Flora of the San Joaquin Desert: Niche, Competition & Floral Mosaics on a Novel Substrate Emma FRYER¹, Ryan O'DELL² and Nishanta RAJAKARUNA^{1,3}</p> <p>1. <i>Department of Biological Sciences, California Polytechnic State University, USA</i> 2. <i>Bureau of Land Management, United States Department of the Interior, USA</i> 3. <i>Unit for Environmental Sciences and Management, North-West University, South Africa</i></p>
S7.P5	<p>Violets of the Allchar arsenic-thallium deposit (Republic of North Macedonia) – proof of hyperaccumulation Ksenija JAKOVLJEVIĆ¹, Tomica MIŠLJENOVIC¹, Katerina BAČEVA ANDONOVSKA², Guillaume ECHEVARRIA^{3,4,5}, Alan J. M. BAKER^{3,4,5,6}, Dennis BRUECKNER⁷ and Antony VAN DER ENT^{3,4,5,8}</p>

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Session 1 - Geology, soils & biogeochemical cycles



A newly-forming ecosystem in a mined ultramafic environment in Barro Alto, Goiás State, Brazil

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ABSTRACT

Ultramafic soils of Barro Alto, Goiás State, Brazil (S14° 58' 6"; W048° 55' 12"), are rich in Fe, Mg, and some heavy metals, such as Cr, Ni, and Co, that are potentially toxic to most plants. Despite these limiting conditions, many plant species grow naturally in these areas. Therefore, they are therefore strong candidates for reclamation of environments degraded by Ni mining. Little is known about how soil parameters change during the early stages of revegetation dynamics on newly restored Ni-mining spoils, particularly in tropical regions (Alday et al., 2012). Characterization of soils from mining spoil heaps showed strong chemical and physical heterogeneity, originated from their distinct mineralogy. There were negligible contents of organic matter (OM), low levels of Ca, P, and K, high bioavailability of Mn, and Cr⁶⁺ that are potentially toxic to the environment, and low microbiological activity (by enzyme activity assessment). Our objective was to evaluate changes in soil and in floristic and microbiological composition in an experiment on the revegetation of mine spoils. Thirteen native herbaceous-shrubs species were selected based on their characteristics of abundance, ability to colonize altered environments and to tolerate or accumulate high amounts of toxic elements. The experiment consisted of hand-sowing seed cocktails composed of 13 native species (NAT), three soil conditioners leguminous (SCL) species, a mixture of NAT+SCL, and no seeding (CONT), which were distributed in 12 permanent plots (0.83m² each). We monitored the soil and vegetation changes in those plots for six years. We monitored the short-term interactions of changes in soil chemical properties, microbiological activities, and plant species succession (composition and structure) in these newly-forming ecosystems. Soil variables showed a general trend of increasing soil OM and pH over time, whereas P and bases saturation (Ca²⁺, Mg²⁺, and K⁺) showed negligible change. Due to the succession of plant species that established themselves in those plots, there were an increase of OM, a four-fold increase in the activity of β -glucosidase and arylsulfatase enzymes, nutrient cycling, and a reduction in the levels of Cr⁶⁺ content in the substrate of more than 80%. The chemical (mainly related to the accumulation of OM and pH increase) and microbiological changes promoted by the rhizospheric activity of native plants started the process of formation of living soil. Our results suggest that the soil-forming material was sufficiently beneficial for plant development. The floristic compositional differences among treatments were mainly driven by a short-term combination of abiotic and stochastic factors. The evolution of this ecosystem allowed the revegetation of an environment that was once totally inhospitable. Based on this information, we developed a methodology for propagation of selected species for planting in field conditions and produced a protocol to re-vegetate mining spoils heaps using native species of ultramafic soils. They will favor the substitution of exotic species commonly present in the seed cocktails used by most of mining companies in this revegetation process.

KEYWORDS: *Cerrado native species, hexavalent chromium, mine spoil materials, soil enzyme activities, ultramafic area.*

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Characterization of chromium-bearing minerals in incipient and highly-weathered serpentine soil profiles in Taiwan

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ABSTRACT

Geogenic Cr in serpentine soils is from Cr-spinels and silicates [1]. Cr-spinels, such as magnetites and chromites, have been considered to be strongly resistant to weathering, while labile Cr is mainly from silicates [2]. However, spinels also contributed to available Cr in serpentine soils, particularly in humid tropics [3]. Thus, the characteristics of Cr-spinels remain unclear for the knowledge of Cr availability in the pedogenetic processes of serpentine soils. The Cr-bearing minerals have distinct magnetic properties. Magnetites are ferrimagnetic with strong magnetism, whereas chromites are paramagnetic with weak magnetism. On the other hand, silicates are commonly nonmagnetic. We collected soil horizon samples from incipient and highly-weathered serpentine soils in eastern Taiwan. The air-dried soils were tested by a portable magnetic susceptibility meter (KT-10, Terraplus (Georadis)) for their magnetic susceptibility (K, SI unit), which is the ratio between the applied magnetic field and the induced magnification. Moreover, the soil samples were separated by applied magnetic field (B) produced by a wet magnetic separator. During the processes, magnetic fractions were obtained under different B: strong magnetic (SM) fractions (B = 1157 G) and weak magnetic (WM) fractions (B = 4650 G). Finally, the remaining were collected as nonmagnetic (NM) fractions. Regarding weight proportion (%), the NM fractions were about 92% in the bulk soils instead of SM (3%) and WM (4%) fractions. However, the proportions of NM fractions gradually decreased toward the surface soils. Additionally, the proportions of SM and WM fractions in the incipient pedon were higher than those in the highly-weathered pedon. The K values were higher in the surface soils than in the subsoils in the two pedons. Nevertheless, the incipient pedon generally exhibited higher K values ($2.65\text{--}5.65 \times 10^{-3}$ SI unit) than the highly-weathered pedon ($1.95\text{--}2.92 \times 10^{-3}$ SI unit). As for the elemental composition, Cr concentration in the SM fractions were the highest, ranging from 6252 mg kg⁻¹ to 30 g kg⁻¹, followed by the WM fractions (639 – 2898 mg kg⁻¹) and NM fractions (453 – 1426 mg kg⁻¹). Summarily, the SM and WM fractions containing magnetites and chromites were corresponding to the low K values in the highly-weathered serpentine soils, indicating that the pedogenesis caused to change in the composition of element in spinels. Hence, the research needed in the future is confirming the spatial distribution of elements and leaching potential of Cr in these Cr-bearing minerals to identify the individual source contribution of geogenic Cr from serpentine soils.

KEYWORDS: *chromium, wet magnetic separator, magnetite, chromite, pedogenesis*

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Ni way! The tricky Ni-biogeochemical cycle in soils under hyperaccumulator of New Caledonia

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ABSTRACT

Ultramafic (UM) soils are particularly challenging for plant growth and are often covered by a distinctive and highly endemic vegetation. At particular, these soils are poor in essential plant nutrient and have high concentrations of metals such as Fe, Mn, Co, Cr or Ni up to the percent level [1]. Among the adaptive physiological strategies characterizing plants on these soils, metal hyperaccumulation leads to the preferential uptake of metal at the soil-plant interface and the transfer of metal from the roots to the leaves, through sap and latex. The Ni-hyperaccumulator *Pycnanthus acuminatus*, an endemic species of New Caledonia, is one of these species, and Ni concentrations in its latex can reach up to 26 wt.% (dry mass) while most plants contain less than 15 ppm (dry mass) of Ni in their aerial organs [2]. This physiology is also suspected to redistribute organic Ni to the soil via the leaf litter degradation and increasing Ni-bioavailability in the soil's surface [3, 4]. Such physiological transfers and processes are suspected to print the $\delta^{60}\text{Ni}$ signatures in the different compartments of the soil and plants. But the mechanisms and extent of Ni fractionation in UM soils due to Ni-hyperaccumulators are not clearly understood and still under debate. The present study focuses on soil profiles developed under hyperaccumulators and non-hyperaccumulators, *P. acuminatus* and its close related *P. fastuosus* respectively, to unravel the effect of hyperaccumulators on the Ni biogeochemical cycle. Ni content within the litter and soils developed under *P. acuminatus* are significantly higher than in profiles developed under *P. fastuosus*. In addition, the bioavailable Ni within soils developed under hyperaccumulator is also higher compared to those developed under *P. fastuosus*, especially in the uppermost horizons, which suggests a non-negligible effect of the hyperaccumulation processes and the litter degradation in the Ni biogeochemical cycle. Coupling the Ni-speciation and Ni isotope ratios in the soil will help us to shed light on biological processes leading to Ni isotope fractionation and highlights the influence of Ni-hyperaccumulators on the Ni biogeochemical cycle in the soil-plant interface.

KEYWORDS: Ultramafic soils, Nickel biogeochemical cycle, Ni-isotope, Hyperaccumulator, New Caledonia

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Nickel and chromium speciation in a small ultramafic catchment: implications for elemental bioavailability to freshwater organisms

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ABSTRACT

In waters draining ultramafic catchments, concentrations of nickel (Ni) and chromium (Cr) naturally exceed the corresponding environmental quality standards or guidelines (EQS or EQG). Such high concentrations of Ni and Cr are of natural origin and ultramafic environments support several species of adapted organisms. However, ultramafic minerals are also actively mined for Ni, a key element in green energies and technologies, or investigated as substrates for CO₂ sequestration. These uses may expose previously unacclimatized biota to potentially harmful levels of Ni and Cr. Identifying the links between Ni and Cr speciation and bioavailability is therefore of critical importance to ensure an environmental friendly use of ultramafic resources. Examination of long-term monitoring data from a small creek flowing through serpentine bedrock (Pluhuv Bor creek [1], Czechia) showed that total filterable Ni concentrations were consistently above the European EQS of 4 µg/L (after bioavailability correction). Similarly, total filterable Cr levels exceeded the EQG proposed by the World Health Organization: 4 µg/L for Cr(VI) and 10 µg/L for Cr(III). Combined chemical and biological investigations were therefore carried out to understand if Ni and Cr bioavailability in Pluhuv Bor waters could be high enough to cause ecotoxic effects in organisms previously unexposed to Ni and Cr. Field work was carried out in the Pluhuv Bor creek in Oct/Nov 2019 (baseflow), Nov/Dec 2021 (minor flood due to snowmelt) and Sept 2022 (baseflow). Elemental speciation was studied using *in situ* Diffusive Gradients in Thin Films, filtration (0.22µm) – ultrafiltration (3kda) and ion-chromatography ICP-MS. The potential combined bioavailability of Ni and Cr was evaluated using standard tests with the model freshwater alga *Raphidocelis subcapitata* and the model crustacean *Daphnia magna*. Total filterable concentrations (all sampling periods) ranged from 33 to 194 µg/L for Ni and 1.9 to 23 µg/L for Cr with the highest values recorded at high the highest flows. Sample fractionation by filtration–ultrafiltration (2021 and 2022 campaigns) showed that ultrafiltered Ni and Cr concentrations did not vary with flow, suggesting that increases in total filterable concentrations at higher flows were caused by an increase in the corresponding colloidal pools. DGT-labile concentrations were in reasonable agreement with ultrafilterable ones for both elements. In the case of Cr, DGT measurements suggested that ultrafilterable Cr mostly occurred as Cr(VI). These results were confirmed by IC ICP-MS analysis for samples collected in 2021. Filtered waters never elicited adverse responses from *R. subcapitata* or *D. magna*, suggesting a limited bioavailability of Ni and Cr. We surmise that the low Ni and Cr bioavailability are linked to the increase in their colloidal pools at high flows.

KEYWORDS: *filtration-ultrafiltration, diffusive gradients in thin films (DGTs), ecotoxicity, colloidal carrier phases, ion-chromatography ICP-MS*

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Rhizosphere processes and nickel mobilization in the nickel hyperaccumulator *Odontarrhena chalcidica*

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ABSTRACT

Nickel hyperaccumulation is a rare phenomenon whereby plants accumulate Ni in aboveground biomass exceeding concentrations of 1000 $\mu\text{g g}^{-1}$ in dry weight [1, 2]. Hyperaccumulator plants targeting Ni commonly occur on ultramafic soils, which are characterised by low nutrients and extreme concentrations of certain metals such as Ni, Cr, and Co. The hyperaccumulation trait makes these plants particularly good candidates for phytomining, in which hyperaccumulators are planted to extract metals from soils and recover them from biomass. However, whether hyperaccumulators are capable of mobilizing larger Ni pools than non-accumulators is still debated. Furthermore, rhizosphere processes of hyperaccumulators are still largely unknown. Especially the biogeochemistry of Fe, P and Ni is assumed to be tightly linked in ultramafic soils since Fe oxides are important Ni and P bearing phases. To investigate the influence of labile and total Ni concentrations in ultramafic soils upon Ni hyperaccumulator plant responses, a gradient of six soils with pseudo-total Ni concentrations ranging from 552 to 1465 mg kg^{-1} and labile (DTPA-extractable) Ni from 41.6 to 158 mg kg^{-1} was created by soil mixing and compared to an additional high pseudo-total (1613 mg kg^{-1}) but low labile (53.1 mg kg^{-1}) Ni soil. The Ni hyperaccumulator *Odontarrhena chalcidica* was grown for 71 days in pots and pore water sampled four times during the experiment to monitor changes in soil solution ionome, pH, and dissolved organic carbon (DOC) content. Labile Ni and Fe fractions as well as P bioavailability were determined before and after the experiment to investigate plant-induced changes. Results showed that pore water Ni and Fe concentrations were significantly increased by *O. chalcidica* as well as DOC concentrations, pore water, and soil pH compared to unplanted soils. Labile Ni was decreased as a result of excessive plant uptake of the labile fraction and perhaps a reduction due to soil alkalinisation. A positive correlation between Ni in shoots and pseudo-total concentrations and pH in soil was observed, although plant Ni concentrations did not clearly show the same linear pattern with soil available Ni. Increased DOC concentrations in pore water were the first indications of root exudation for *O. chalcidica* and presumably promoted enhanced Ni and Fe solubilization in soil pore water. Our results showed a clear root-induced Ni and Fe mobilization in the rhizosphere of *O. chalcidica* and suggested a nutrient uptake mechanism based on soil alkalinization and exudation of organic ligands.

KEYWORDS: *hyperaccumulation, rhizosphere, mobilization*

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Stocks and biogeochemical cycling of soil-derived nutrients in an ultramafic rain forest in New Caledonia

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ABSTRACT

Ultramafic rain forests in New Caledonia evolved on some of the most nutrient impoverished soils globally and are some of the slowest-growing tropical forests known. This study aimed to determine nutrient stocks and elucidate the biogeochemical cycling of nutrients in a remnant lowland rain forest in southern New Caledonia. Based on an inventory of a 1-ha permanent plot, exhaustive plant tissue sampling was undertaken of all large trees (diameter at breast height ≥ 15 cm) in a 0.25-ha subset of the plot in tandem with collecting 100 soil samples. All samples were analyzed for major nutrient concentrations and the results show that most of the magnesium was contained in the soil (96.9%), whereas a large fraction of calcium (46.5%) and phosphorus (16.0%), and the majority of potassium (81.5%) were contained in the standing biomass. This study has revealed how tightly these soil-derived nutrients are cycling in this system. Ultimately, this information will be essential for efforts to restore rain forest in New Caledonia, where the biomass (and contained nutrients) has been removed.

KEYWORDS: *Nutrients, Ultramafic, Biogeochemistry, Potassium, Phosphorus*

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Studies on arbuscular mycorrhizal fungi in New Caledonian ultramafic soils: A synthesis

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ABSTRACT

If the knowledge about New Caledonian serpentine ecosystems has increased greatly during the past half-century, research on soil microflora and plant symbionts started only in the nineties and was mainly published during the last two decades. Here we synthesize the studies on arbuscular mycorrhizal fungi (AMF), focusing particularly on ecological and restoration aspects. Research on AMF consisted firstly of a global and inventory approach aiming to produce basic but essential lacking knowledge. These studies showed that AMF are abundant in ultramafic soils and concerned nearly all plant species of these ecosystems. Even Ni-hyperaccumulator plants and sedges, generally considered non-mycorrhizal, were found to be functionally colonized by AMF in New Caledonian ultramafic soils. The adaptation of AMF communities to the extreme conditions of these soils led to high levels of metal tolerance (particularly to Ni) and noticeable originality of the taxa. Several new species were described. A relatively high AMF diversity was found in the studied serpentine ecosystems. The influence of these symbionts on plant growth and adaptation was assessed in greenhouse and field conditions. An accurate selection of AMF isolates that improve plant growth, and plant metal tolerance was performed. It was demonstrated that combinations of AMF isolates with complementary functional traits showed high synergistic effects on plant development. Finally, a partnership with a biotechnological company led to the production of an efficient commercial inoculant now used in the ecological restoration of mine-degraded areas. Today studies are focused mainly on the additive effects of AMF and mycorrhiza-helper bacteria.

KEYWORDS: *AMF; serpentine ecosystems; abiotic stress; toxic metals; restoration ecology; New Caledonia*

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Ancient soils in non-glaciated mountain areas on peridotite: the Lanzo Ultramafic Massif example (TO, NW Italy)

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ABSTRACT

The Lanzo Ultramafic Massif (LUM) is one of the largest peridotite outcrops in the world; the peridotitic rocks are only slightly serpentinized, and they are often deeply weathered, and they are substituted by antigoritic serpentinite only locally, in the westernmost part of the massif. Given the low elevation, the LUM was never glaciated during the Quaternary, and the surface characterized by extremely well-developed fossil periglacial features, such as blockstreams, blockfields and large-scale patterned ground (Paro, 2011). These periglacial landforms have an age ranging the whole Quaternary, while Pliocene sediments are observed close to the base of the massif (Forno et al., 2010). The vegetation is sparse, and rich in endemic serpentine species, some of which are Ni hyperaccumulators. Inside and near the ancient landforms, extremely well developed soils are found. In particular, deep Podzolic soils are often found below 1-2 m-thick blockstreams in the highest slopes, above 1000 m a.s.l., characterized by thick E and cemented Bsm horizons; at lower elevations, soils between blockstreams are characterized by layers with increasing pedogenic indicators with depth (clay coatings, rubification and fragipan formation), with the deepest horizons strongly hardened by fragipan and deeply rubified. In the stream incisions, it is possible to observe red, strongly cemented horizons likely developed during the Pliocene, under subtropical conditions, partly resembling lateritic materials. The Fe and heavy metal concentrations are extremely high, and it is a strong ecological constraint even in these ancient soils.

KEYWORDS: *paleosols; heavy metals; Plio-Pleistocene.*

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Drivers of Cr and Ni Mobility in a Serpentine Catchment

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ABSTRACT

Hydrochemical changes between 1991 and 2022 were assessed based on weekly sampling of streamwater at Pluhův Bor (PLB) in western Bohemia, Czech Republic. This small catchment (21.6 ha) is underlain mainly by serpentinite, with occasional veins of tremolite schists, actinolite schists and blocks of amphibolite. It is forested mainly by Norway spruce (*Picea abies*) with a small proportion of Scots pine (*Pinus sylvestris*) [1]. PLB exhibited one of the most dramatic decreases in streamwater sulfate concentrations ever recorded [2]. The annual discharge-weighted mean of sulfate concentrations declined from 50 mg/L in 1992 to 10 mg/L in 2002 (-3.6 mg/L/yr) because of the desulfurization of lignite burning power plants in the region. A new steady state with respect to sulfate concentrations was observed between 2010 and 2022, with concentrations only between 6 and 9 mg/L. Steady increase of annual discharge-weighted mean dissolved organic carbon (DOC) concentrations in streamwater was observed at PLB (+0.8 mg/L/yr, $P < 0.001$) for the entire period of observations due to declining ionic strength of precipitation and drainage water, and thermally moderated increased dissolution of soluble soil organic matter pools [3]. Atmospheric deposition of Cr and Ni at PLB was negligible compared to large sources of geogenic nickel [1] and chromium [4] in bedrocks and soils at PLB. Discharge-weighted mean streamwater concentrations were 30 µg Cr/L (± 3 µg/L) and 156 µg Ni/L (± 27 µg/L), almost twice higher than simple mean values, thus reflecting a positive correlation of these metals with discharge. Annual streamwater fluxes were 73 ± 19 g Cr/ha/yr and 419 ± 150 g Ni/ha/yr. Standard dissolved fraction in streamwater, filtered by a membrane < 0.45 µm, was 91% and 95% of total unfiltered Cr and Ni, respectively. However, a recent study performed in 2021-2022 using ultrafiltration (< 3 kDa) showed that truly dissolved fractions of Cr and Ni were much smaller with potential implications for ecotoxicity (Vignati et al., this volume). A slight increase of long-term Cr and Ni concentrations was observed at PLB. The positive correlation of DOC, Cr and Ni streamwater concentrations in short-term (time scale of hours) is due to water flow paths routed through the organically rich surficial horizons during high-flow events mobilizing carrier phases such as DOC or colloidal iron. This contribution was supported by the CSF (21-22810J), CGS (310690), eLTER PLUS and Fulbright.

KEYWORDS: forest catchment, long-term monitoring, streamwater, dissolved organic carbon, sulfate, chromium, nickel, filtered and unfiltered concentrations, element fluxes, hydrological events

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The Nickel Biogeochemical Cycle in a Tropical Hyperaccumulator Soil-Plant System

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ABSTRACT

Nickel (Ni) hyperaccumulator plants play a major role in Ni recycling in ultramafic ecosystems, which may be more pronounced in a wet tropical environment. In this study we present the biogeochemical cycling of Ni (and other elements) on an ultramafic Cambisol by a tropical Ni hyperaccumulator, *i.e.*, *Phyllanthus rufuschaneyi*, one of the most promising species globally for agromining. The objectives were: (i) to study the biogeochemical cycling of a natural stand of *Phyllanthus rufuschaneyi* in order to assess and to evaluate the natural fluxes of Ni and other elements in the ecosystem; (ii) to manipulate such an ecosystem in order to perform a sensitivity test of the ecosystem for the following flux of litter returning to the soil. Two parallel stands of *P. rufuschaneyi* were instrumented, monitored and compared over two years (2018 and 2019), (i) a natural secondary 100-m² forest and (ii) a densely planted experimental field, in which litter returns to the soil were calibrated; from no return (export) to a doubling of the return (*i.e.* 0, 1 and 2 kg m⁻² per annum).

This study did not confirm allelopathy of tropical hyperaccumulator plants, despite the extreme influence of Ni hyperaccumulators in building up considerable available Ni stocks in topsoils. Results also confirmed rapid lixiviation of Nickel from soils by natural waters despite plant recycling. Overall, nickel cycle was mainly driven by internal fluxes, *i.e.* litter decay, degradation and recycling of the hyperaccumulator biomass. The percentage of Ni recycled by litterfall tended to decrease with increasing litter addition to the soil and was not influenced by coppicing, at least in such a short term. Nickel turnover should be taken into account when designing tropical agromining crops and natural secondary forests are a good surrogate to assess the long term impacts of agromining (e.g. soil nickel depletion). Results suggested a significant and steady contribution of mineral weathering to Ni available pools in the soil-plant system. Further study of the weathering processes would help to specify the contribution of bedrock and soil mineral horizons in the Ni and nutrient budgets of the system and the rates of rock weathering.

KEYWORDS :

biogeochemistry; agromining; litter; nutrient turnover

Application of clay minerals in remediation of heavy metal pollution in soil

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ABSTRACT

Soil pollution is the most important environmental issues causing serious problems to humans and all living creatures. The problem of removing of heavy metals such as cadmium, lead, nickel, chromium, iron, zinc and copper from soil is an important process and is becoming more important with the increasing of industrial activities. Clay minerals have attracted much attention for metal stabilization due to their high specific area, liming effect, excellent ion exchange capacity and abundant surface hydroxyl groups, strong mechanical stability, stable chemical properties, lower cost and environmentally friendly. Bentonite is used as an adsorbent for removal of metal ions because of its cation exchange capacity, larger surface area and adsorptive capacity for different organic and inorganic ions. The aim of this study was to evaluate the adsorption ability of natural soil components such as clays to the remediation of industrial contaminated soils in Albania. We selected for study, the Bentonite of the Prrenjas area (south-east of Albania). Adsorption experiments carried out using metal solutions of Cr, Ni, Zn, Fe extracted from soils polluted by industrial activities in Albania. For extraction of Clay we used batch technique. We study: effect of contact time on adsorption of metals ions; effect of pH on adsorption of metal ions; impact of the amount of adsorbent on adsorption of metal ions; effects of initial metals concentration. The results suggested us the application of clays to the contaminated soils for remediation.

Assessing ecological, environmental, and human health risks associated with ultramafic soils in Brazil

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ABSTRACT

Ultramafic soils can pose ecosystem and human risks due to high levels of heavy metals (mainly Cr, Ni, and Co). Soil chemical and mineralogical attributes control the mobility of these elements. In the tropics, weathering conditions can intensify the mobility and availability of these metals to living beings. Therefore, environmental and human health risk assessments are necessary for ultramafic environments. In this scenario, our objectives were to characterize Brazilian ultramafic soils and evaluate the contents and mobility of Cr, Ni, and Co in soils to estimate environmental, ecological, and human health risks from exposure to ultramafic soils. We carried out chemical and mineralogical characterizations as well as the total, pseudo total, available, bioaccessible (gastric, pulmonary, and dermal), and soil fractions analyses of Cr, Ni, and Co in two ultramafic soils from different regions of Brazil (Niquelândia – Goiás state and Buenos Aires – Pernambuco state). Additionally, the carcinogenic and (non)-carcinogenic risks were estimated. The mean total metal concentrations (mg kg^{-1}) for the Niquelândia and Buenos Aires soils, respectively, were Co (373.5 and 349.2), Cr (1844.5 and 2485.5), and Ni (9597.5 and 1428.5), respectively. The available concentrations (mg kg^{-1}), in turn, were Ni (220.9 and 66.1), Co (2.4 and 1.0), and Cr ($< \text{LD}$). The soil fractionation showed that Fe and Mn oxides were the main pools for Co in both soils and Ni in the Buenos Aires soil, whereas Cr in both soils and Fe in Niquelândia soils were mostly retained in residual fractions. No environmental risk was associated with Co and Cr in the soils, but low (1.1 %) and medium (13 %) environmental risks regarding Ni were found for the Niquelândia and Buenos Aires soils, respectively. On the other hand, ecological risks were estimated high for the Buenos Aires soil ($\text{PERI} = 522.8$) and significantly elevated in Niquelândia ($\text{PERI} = 1759.9$). The metals showed low pulmonary, gastric, and dermal bioaccessibility; hence, the carcinogenic and non-carcinogenic risks were acceptable. The high contents of iron and manganese oxides in soils are likely responsible for metals' low availability and bioaccessibility. The high ecological risks found suggest that the microbial communities in these environments are adapted to metalliferous conditions and that the erosion and transport of these soils can damage microorganisms elsewhere.

KEYWORDS: *bioaccessibility, heavy metals, serpentine soils.*

Distribution of Heavy Metals in the Different Compartments in Lake Dushku, Albania

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ABSTRACT

Lake Dushku is a natural eutrophic lake located in the base of Komjan mountain, Albania, at about 1200 m asl (40°48'0.45"N, 20°19'45.48"E). Its total surface is 0.4 km². The substrate of this area is dominated by serpentine rocks, formed during Oligocene. Lake Dushku is surrounded by a forests belt, dominated by *Pinus nigra* Arn. Helophytes such as *Phragmites australis* (Cav.) Trin. ex Stendel, *Typha angustifolia* L., *T. latifolia* L., *Scirpus lacustris* L., *Heleocharis palustris* (L.) Roemer & Schultes, *Juncus inflexus* L., and *Iris pseudacorus* L., are the main components of the vegetation. They form stands or belts of different width in the lake, frequently accompanied by the floating hydrophytes *Nymphaea alba* L. and *Nuphar lutea* (L.) Sibth. & Sm. Heavy metal concentrations in water, sediment, *Nymphaea alba*, were investigated at 12 selected sites and in the surrounded area. The surface water was found to have low levels of heavy metals (10 µg/l), but the sediment contained very high levels with concentrations reaching 2750 mg/kg for Ni, 670 mg/kg for Cr, 110mg/kg for Co. DTPA extraction of metals demonstrates that the available heavy metals in the sediment are high (400 mg/kg Ni). Heavy metals were found in plants (stem, leaves and flowers of *Nymphaea alba*).

KEYWORDS: Heavy metals, · Lake Dushku, ·serpentine, · DTPA extraction

Interactions of Cr(III)/Cr(VI) Mixtures with Freshwater Algae: a Stable Isotope Approach

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ABSTRACT

Trivalent and hexavalent chromium have contrasting environmental behavior, bioavailability and ecotoxicity. Current consensus considers Cr(VI) as the most toxic form, but studies documenting non-negligible bioavailability and ecotoxicity of Cr(III) are increasing. Furthermore, mixtures of Cr(VI) and Cr(III) are usually found in natural environments. Apart from two seminal studies published in 2022 [1,2], the bioavailability and ecotoxicity of Cr(III)/Cr(VI) mixtures are virtually *terra incognita*. Ecotoxicity testing of Cr(III)/Cr(VI) mixtures was carried out using the model green alga *Raphidocelis subcapitata* following standardized procedures. Algae were exposed to Cr(III), Cr(VI) and Cr(III)+Cr(VI) mixtures using solutions isotopically enriched in ⁵⁰Cr(III) and ⁵³Cr(VI). Absence of Cr(III)/Cr(VI) redox interconversions was previously verified [3]. Concentrations causing a 50% decrease in algal growth (EC₅₀) were about 60 µg/L for Cr(III) and 100 µg/L for Cr(VI), meaning that Cr(III) was more toxic than Cr(VI). The combined effects of Cr(III)+Cr(VI) mixtures followed the response addition model without evidence of synergisms or antagonisms between the two valence states. According to a three dimensional surface model of algal response, Cr(III) and Cr(VI) had the same effect on algal growth at a concentration of 57 µg/L. However, filterable Cr(III) concentrations decreased by more than 90% during the test. The actual Cr(III) mean concentration was therefore estimated at 13 µg/L, confirming that Cr(III) was more toxic than Cr(VI). Analogous experiences were carried out in a test medium amended with 0.4 mg/L goethite, a well-known colloidal carrier phase in natural waters. The individual EC₅₀ of Cr(III) and Cr(VI) in the presence of goethite were similar to those in the standard test medium. In experiments with a ternary mixture of Cr(III), Cr(VI) and goethite, Cr(III) was more toxic than Cr(VI) and no interactions were identified between chromium and goethite. A third round of tests was performed in filtered waters (0.22 µm) from a small ultramafic creek (Pluhuv Bor, Czechia) containing colloidal Fe and organic carbon. In these waters, Cr(VI) was found to be more toxic than Cr(III), suggesting a strong role of organic matter in regulating Cr(III) and Cr(VI) ecotoxicity to algae.

KEYWORDS:

mixture ecotoxicity; colloidal carrier phases, ultramafic waters; stable isotopes

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The influence of fire and fire retardant (Phos-chek®) on plant diversity and non-native species abundance in California's serpentine chaparral

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ABSTRACT

Fire and harsh soils play key roles in maintaining plant communities. However, the effects of fire-retardant on plant diversity and soil geochemical dynamic on serpentine soils are unclear. We investigated how fire and a nutrient-rich fire retardant, *Phos-chek* (PC), influence diversity and the relative abundance of native and non-native species on serpentine soils. Our study area consisted of serpentine chaparral which burned in 2017 and was treated with PC. Treatments consisted of burnt serpentine, unburnt serpentine treated with PC, and unburnt serpentine as the control. Soil chemistry and plant species cover were assessed yearly for two years beginning 6 months after the fire. We tested the effect of PC and fire on diversity and non-native abundance using generalized linear mixed-effect models, tukey pairwise comparisons, and the likelihood-ratio test. We found no significant effect of burning or PC application on diversity of species. However, non-native species abundance was significantly different among treatments directly following the fire, with complete elimination of non-native abundance in the burned treatment, and an increase in non-native abundance in the PC treatment. In subsequent years, non-native abundance returned to levels found in the unburnt treatment, increasing on burnt serpentine and decreasing on unburnt serpentine treated with PC. We explore the theoretical geochemical feedback potentially causing the underlying patterns on unburnt PC treatments through the reaction and fates of the trioctahedral layer. This study helps better predict and plan for shifts in species distributions in response to fire and PC in serpentine chaparral.

KEYWORDS: *fire, fire retardant, biodiversity, biogeochemical, serpentine, chaparral*

Wettability of serpentine and other minerals upon rising temperatures

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ABSTRACT

The North-Western Italian Alps are characterized by occurrence of serpentinite rocks¹ and are not unfamiliar to fire occurrence². Both these aspects affect soil and above-ground vegetation^{3,4}, originating landscapes with specific and unique features. Mountain serpentine regions are potentially prone to after-fire erosion phenomena due to a marked steepness degree. Run-off and erosion are tightly related to the extent of soil water repellency (WR)⁵. Water repellency often increases after fire, but is regulated by a variety of factors; among them, the contribution of the parent-material mineralogical composition to the actual soil WR remains widely obscure, despite the silicate structure is expected to affect mineral-water interactions⁶. Four pure silicate minerals (serpentine, smectite, kaolinite, biotite) were combined with two litter types (*Fagus sylvatica* L., BEECH, and *Pinus sylvestris* L., PINE) in different proportions, so as to originate mixtures containing growing organic carbon (OC) contents (10-30-70 g kg⁻¹). The mixtures were heated in a T-range from 100 to 300 °C and characterized. We measured WR by Water Drop Penetration Time (WDPT) test, specific surface area (SSA), pH, and OM content and composition (by FT-IR and ¹³C NMR). At room temperature, all silicates were wettable, and WR increased with increasing OC loads. No univocal differentiation upon OM type (BEECH vs. PINE) emerged for smectite-mixtures, while WR induced by pine was twice as much that of beech in the case of serpentine. WR was maximized at 200 °C and drastically lost above this T. Mineral mixtures generally exhibited a higher WR in presence of greater OC loads, but no clear differences between the two types of litter emerged. The OM characterization revealed that, albeit deriving from two distinct forest covers, the composition of the litter types followed similar heat-induced transformations. Serpentine showed comparable WR upon heating to kaolinite and biotite (WDPT>8.000 s at 200 °C), while smectite samples systematically displayed the lowest WDPT values, ca. 200 s at 200 °C. This suggests that the interaction between mineral phases and OM is primarily ruled by the surfaces available for bond creation (SSA: 4-13 m² g⁻¹ vs >400 m² g⁻¹) and, only secondly, by the specific wettability of these surfaces (2:1 vs. 1:1 phyllosilicates). The mechanistic understanding on organo-mineral associations at the lab-scale revealed that even modest OM contents (≤30 g kg⁻¹) would be sufficient to trigger WR in serpentine soils after heating and, thus, a similar behaviour is to be expected at larger scales in natural fire-affected Alpine areas.

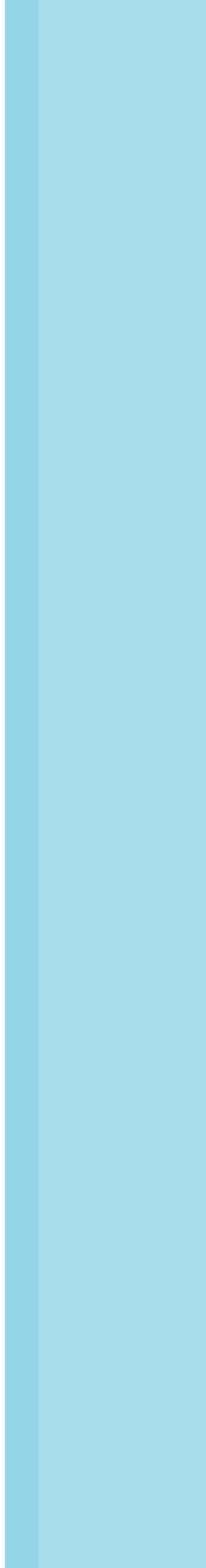
KEYWORDS: water repellency; thermal treatment; organic compounds; phyllosilicates; forest fires

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Session 2 - Biodiversity: taxonomy and systematics



Additions to the lichen flora of Albania

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ABSTRACT

Albania is characterized by several natural conditions suitable for a wide range of lichen species, as occurs along the Balkan Peninsula. Despite this, it is still one of the least lichenologically investigated countries in Europe. Its long isolation in the 20th century and the lack of resident lichenologists have contributed to limit sources of lichen diversity information about the country. Over the last 20 years, Hafellner [1] has provided a checklist of lichens based on historical records by several authors, such as Markgraf [2], and data collected by the colleague L. Kashta during some fieldwork on higher plants, resulting in a lichenological publication [3]. Since then, single species or additional records have been added by other authors [as for example in 4]. After these publications, a greater number of epiphytic and epilithic lichens have been reported, however, considering the size of the country, the high habitat diversity, and the high-altitude gradient (0-2764 m) that characterize its territory, there is still much to discover about lichen diversity and distribution in Albania. This is especially true for epilithic and terricolous lichens that colonize ultramafic habitats. In fact, serpentine rocks and soils have so far been lichenologically scarcely explored, despite being rather common in the country, probably due to their difficulty in being reached. Among the publications cited above, only two authors report data from samples collected on serpentine, for a total of about 46 taxa [3,4]. During our recent field excursions focused on metallophytes, several lichen specimens were collected across different ultramafic areas of Albania. Most of the visited areas have never been covered for lichenological studies: rock pastures and rocky scrub margins at Qafe Shtamë (Krujë district), overhanging cliffs of serpentine along Devoll river valley, pastures and stony ground on ultramafic rocky soil in Vallamarë mountain (Gramsh district), ultramafic rocks and cliffs in Moravë mountain and along Devoll river valley near Lozhan valley (Korçë district), from stones and ultramafic soil rocks from the slag flows of a Cr mine in Pishkash (Librazhd-Perrënjës district). Specimens have been identified using lichenological standard methods, including Thin Layer Chromatography analyses and UV light, and deposited in private herbaria of the first author. More than 100 specimens of lichen have been collected among which there are several species never collected on serpentines and new recorded for Albania before. This work is the first contribution of a larger project that will involve ad hoc sampling in the country in the next few years.

KEYWORDS: *biodiversity; lichen; ultramafic environments; flora of Albania*

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Seed endosphere of *Brassicales* and *Asterales* hyperaccumulating plants harbor a stable core microbiome

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ABSTRACT

Our knowledge of rhizosphere microbial diversity is now well-established [1, 2]. Conversely, little is known concerning the diversity of endophytic bacteria, particularly within seeds of hyperaccumulating plants, despite seed germination being a critical stage in plant life cycle [3]. However, the seed microbiota is currently of great interest in the scientific community. It is now understood that part of the seed microbiome is composed of vertically-transmitted endophytes that may have co-evolved with their host plant, with a symbiotic relationship shared across generations [4, 5]. It is even possible to consider that some endophytic bacteria could be common to the seeds of hyperaccumulating plants, thereby conferring them an evolutionary advantage over non-hyperaccumulating plants when confronted to biotic or abiotic stress. This study focuses on the endophytic bacterial diversity of metal hyperaccumulating and non-hyperaccumulating plants (93 seed samples from Mediterranean regions, Oceania, South-East Asia) including 17 different genus and 45 species. Alpha diversity index readings were low. Furthermore, the difference in composition between the endophyte communities was significantly explained by the botanical family and partly by the hyperaccumulation trait. Thus, a set of 12 OTUs common to seeds of hyperaccumulating *Asterales* and hyperaccumulating *Brassicales* was identified and could be considered as a ‘stable’ core microbiome. This could confer an adaptive advantage on hyperaccumulators when faced with abiotic stresses with the potential expression of PGP traits.

KEYWORDS: *endophytic bacterial diversity, seed, hyperaccumulator*

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Soil contamination levels of nickel imply changes in the bacterial communities associated to the rhizosphere and endosphere of *Odontarrhena chalcidica*

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ABSTRACT

Some plants, called metal hyperaccumulators and endemic to soils naturally rich in metals, reached a high degree of adaptation and are able to accumulate large amounts of metals, such as nickel, in their aerial parts. This was the origin of the development of agromining, a technology based on the culture of these particular plants, such as *Odontarrhena chalcidica*, in order to produce high value metal compounds, such as nickel [1]. Recently, the concept of microbe-assisted agromining had been introduced to underline the role of plant-associated microorganisms, both rhizosphere and endophytic, in metal bioavailability and uptake by host plants [2]. However, the efficiency of plant-associated bacterial communities depends on a complex array of interacting factors, including metal concentration in soil. A better understand of the impact of different soil contamination levels of nickel on the structure and diversity of rhizosphere and endospheric bacterial communities is needed. In this work, we characterized, using high-throughput sequencing, the bacterial communities associated to *Odontarrhena chalcidica* growing in controlled conditions on an ultramafic soil with various levels of nickel contamination obtained by nickel-spiking in the soil. Our results support that an increase of the available nickel in soil induced changes of the dominant bacterial genus in the communities of the rhizosphere soil, but also in the root and shoot endosphere. This increase of available nickel also implied changes in the relative abundance of the predicted functions, particularly for the rhizosphere and root endospheric bacterial communities. In addition, topological features of the bacterial networks seemed to indicate that at intermediate level of nickel contamination, two coexisting bacterial sub-communities were in competition, one adapted to « low » soil nickel content and the other to higher nickel content, while the bacterial communities were more stable at the lowest and the highest nickel soil contamination levels. Finally, our results revealed shifts in the microbial community's structure and functions depending of the gradient of soil nickel availability in the soil.

KEYWORDS: *bacterial diversity, hyperaccumulator, nickel*

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Grassland flora of serpentine areas in North Pindus (Greece)

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ABSTRACT

In Europe, the largest serpentine (ultramafic) outcrops are located in the Balkan Peninsula, presenting a high floristic diversity and speciation. In Greece, the Pindus mountain range due to the presence of large serpentine outcrops, is characterized as a floristic diversity hotspot including a large number of hyperaccumulators which have potential use in agromining. The present work is part of a more in-depth study of floristic composition and ecology of plant communities in serpentine grasslands of the Pindus mountain range that started in 2021 and continues to date. During the field survey, 120 relevés were sampled according to the Braun-Blanquet method at 12 sites. According to the results so far, 493 plant taxa belonging to 61 families have been identified. Asteraceae, Poaceae and Fabaceae are the most abundant families. Of the total number of plant taxa, 20 taxa are Greek endemics, while 55 taxa are distributed in the Balkan Peninsula and 95 taxa in the Mediterranean region. The highest number of plant species are hemicryptophytes (53%) and therophytes (22%). Eight species known as nickel hyperaccumulators (*Centaurea thracica*, *Bornmuellera baldaccii*, *Bornmuellera emarginata*, *Bornmuellera tymphaea*, *Noccaea boeotica*, *Noccaea tymphaea*, *Odontarrhena chalcidica* and *Odontarrhena smolikana*) were found in 109 relevés carried out at all sites, being important members of the existing vegetation.

KEYWORDS: *ultramafic, Balkans, nickel hyperaccumulators*

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Investigating the Diversity and Function of Microorganisms in Serpentine and Non-Serpentine Soils across California

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ABSTRACT

Serpentine soils are known for their high levels of heavy metals and high magnesium-to-calcium ratios which places a strong selective pressure on the plants and microorganisms that inhabit these soils. Although some plants are endemic to serpentine soils, most plants cannot survive on these soils. Our previous research has shown that serpentine-indifferent plants, which are able to thrive on both serpentine and non-serpentine soils, may be partially sustained by their association with locally adapted plant growth-promoting bacteria. The role of microorganisms in supporting plant diversity and other ecosystem services are continuing to be elucidated and characterizing the taxonomic and functional diversity of microbes on and off serpentine soils will further those efforts. The goal of our project was to identify taxa that are associated with either serpentine or nonserpentine soils and characterize the variation in plant-growth-promoting properties of microorganisms isolated from those soils across California. To achieve this, we conducted shotgun metagenomic sequencing and culture-dependent physiological assays. Soil samples were collected from 23 sites across California between November and December 2020, and taxonomic classification was conducted in Kaiju using the microbial subset of the NCBI BLAST non-redundant protein database. Functional analysis was conducted using MEGAN6 Ultimate Edition. After removing contaminants, 41,326 taxa were identified from the metagenome including 36,752 bacteria (89%), 2,881 eukaryotes (7%), and 1,693 archaea (4%). The relative abundance and alpha diversity of these kingdoms are similar across serpentine and nonserpentine soils. Our results show that soil chemistry and site location play a significant role in shaping the bacterial and eukaryotic but not the archaeal communities in both serpentine and non-serpentine soils. According to random forest analysis, some bacterial features include *Mesorhizobium*, *Rhizobium*, and *Pontibacter*; some archaeal features include *Nitrososphaera* and *Natronomonas*; some eukaryotic features include *Melanogaster*, *Serendipita*, and *Pochonia*. Most of these genera are more abundant in nonserpentine soils, but *Nitrososphaera* and *Pontibacter* are more abundant in serpentine soils. Results of preliminary functional analysis show that both serpentine and nonserpentine soils have a Ni/Fe large and small subunit (Hyd1 + Hyd2) and functions related to siderophore production while only serpentine soils had a Ni/Fe metallocenter assembly protein (HypCDE). Bacteria in serpentine soils showed higher levels of siderophore production in the presence of nickel, while bacteria in non-serpentine soils showed higher levels of siderophore production in the absence of nickel. Approximately 50% of the isolated microorganisms from both soil types were capable of fixing nitrogen, while 20% were able to solubilize phosphorus. This study has the potential to contribute to our understanding of nutrient cycling in metal-rich and drought-prone soils and has implications for basic science in plant-microbe interactions as well as practical applications in agriculture and bioremediation. Additionally, the findings of this research will be relevant to conservation policy as they can inform efforts to preserve and protect unique soil ecosystems and the microorganisms that inhabit them.

KEYWORDS: *plant growth-promoting bacteria; soil microbial diversity; local adaptation; kaiju; MEGAN6*

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The checklist of serpentinophytes (serpentine restricted flora) as a tool for researching and conservation in serpentine ecosystems (southern Iberian Peninsula, Spain)

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ABSTRACT

The establishment of a complete ecological-floristic checklist of serpentine flora is essential for the assessment, management, and conservation of serpentine ecosystems. The highly evolved and specialized flora that manages to inhabit these harsh ecosystems includes a very specialized and highly evolved group of plants known as serpentinophytes. Serpentinophytes are linked exclusively or almost exclusively to serpentine ecosystems. The current existing list of obligate serpentinophytes (obligate endemics) which exist in the serpentine ecosystems of the southern Iberian Peninsula (Spain) consists of 22 taxa (Pérez Latorre et al., 2013; Perez Latorre et al., 2018). However, new fieldwork and research using GBIF.ORG have been done resulting in the description of new soil endemics and the discovery of new populations of this specialized flora. The revised list is composed of 24 obligate serpentinophytes, 2 preferential and 4 subserpentinophytes in a total of 430 Km² of serpentine ecosystems in the South Iberian Peninsula outcrops. Serpentinophyte richness per outcrop has been calculated as a function of a logarithm-adjusted area. This updated checklist includes the following data for each serpentinophyte: taxonomical information, type of endemism following the classification of Pérez-Latorre & al. (2013), biogeography, biological type, altitudinal range, vegetation (syntaxon in which it appears), threat category ecology, and hyperaccumulation information. The obtained data can be useful for the management of these valuable ecosystems.

KEYWORDS: *Updated checklist, Spain, serpentinophytes, conservation*

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Bacterial and fungal diversity found in serpentine and non-serpentine soil crusts from two different climatic regions in South Africa

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ABSTRACT

Serpentine soils are characterized by nutrient imbalances and toxic heavy metals and are known to host specialized depauperate plant communities of species with specialized adaptations. However, little is currently known about serpentine microbial diversity and whether it follows similar patterns. Here we investigated the bacterial and fungal diversity of biological soil crusts from serpentine and non-serpentine soils, at different precipitation levels and different average temperatures. The bacterial community was characterized using 16S rDNA metabarcoding and the fungal community by using ITS metabarcoding. A PerMANOVA showed that the microbial composition of serpentine sites differed significantly from non-serpentine sites ($p=0.006$). Fungal diversity was higher at non-serpentine localities than at serpentine localities. Alphaproteobacteria, Actinobacteria and Firmicutes were present at all sites. The fungal class Dothideomycetes was the most diverse group detected at all study sites. The bacterial genera *Azospirillum*, *Bradyrhizobium*, *Candidatus*, *Koribacter*, *Flavisolibacter*, *Gemmata*, *Oenococcus*, *Rhodoplanes* and *Dactylosporangium* as well as the fungal genera *Archaeorhizomyces*, *Leptosphaeria*, *Phaeococcomyces* and *Penidiella* were only found on serpentine soils, but are not considered unique to this type of habitat. It can be concluded that edaphic factors influenced the composition of fungal diversity and a decrease in fungal diversity in serpentine soil crusts was observed.

KEYWORDS: *Serpentine geoecology, biological soil crusts, microbial diversity, fungi, bacteria, metabarcoding*

New *Stylochaeton* species from ultramafic soil in South Africa

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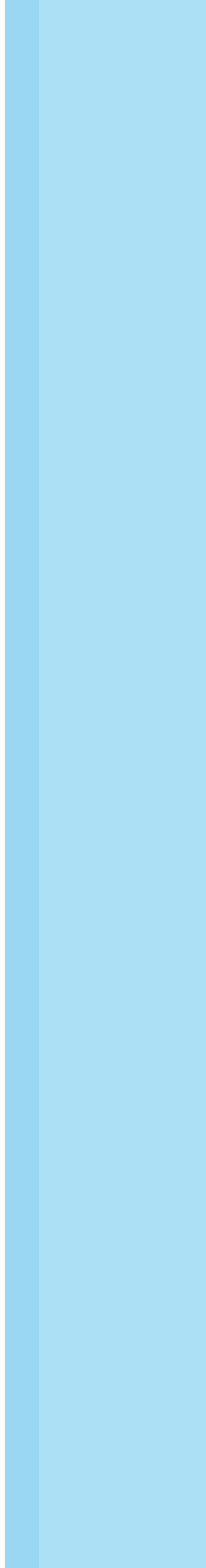
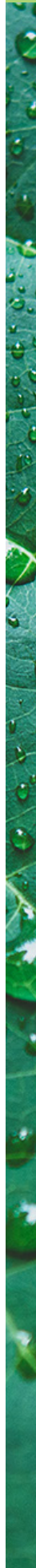
ABSTRACT

Stylochaeton Lepr. is a genus of 15 species in the *Araceae*, distributed across tropical and southeastern subtropical Africa in savannah and humid, deciduous forests. *Stylochaeton natalense* Schott has been the only recognised *Stylochaeton* species to occur in South Africa. It is found in the northeastern regions of South Africa and its distribution extends into Mozambique and further north to Tanzania. During ecological surveys in the Sekhukhuneland Centre of Plant Endemism in the Limpopo province of South Africa, two new species of *Stylochaeton* (*S. glaucophylla* and *S. sekhukhuniense*) were found growing sympatrically with *S. natalense*, although not with one another. These species differ from *S. natalense* mainly in leaf shape, spathe colour and larger, subterranean fruit. The new species are mainly distinguished from each other by their leaf shape and size (the front and basal lobes in *S. glaucophylla* are linear and longer, whereas they are elliptic and shorter in *S. sekhukhuniense*), colour (leaf blades of *S. glaucophyllum* are greenish blue, whereas those of *S. sekhukhuniense* are green), and whether the petiolar sheath is ligulate (*S. sekhukhuniense*) or not (*S. glaucophylla*). The spadix and infructescence grow partly below the ground in both new species. The new species are endemic to pyroxenite hill and mountain slopes (*S. glaucophylla*) or low-lying areas (*S. sekhukhuniense*) of Sekhukhuneland. Although these species show a preference for and are endemic to metalliferous soil, leaf and stem analyses did not show evidence of metal hyperaccumulation. The description of these species is noteworthy, as the ultramafic soil of Sekhukhuneland now seems to be a center of adaptive speciation for two genera in the *Araceae* (the other being *Zantedeschia*).

KEYWORDS: *bushveld arum, endemism, speciation, taxonomy, ultramafic soil*



Session 3 - Ecology & evolution



Community ionomics of a hyperdiverse shrubland: the maquis on ultramafic substrates of New Caledonia

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ABSTRACT

The island of New Caledonia in the southwest Pacific has particularly large surfaces of ultramafic substrates which have low concentration of macronutrients (N, P, K) and excess concentration of metals (Cr, Mn, Ni). It has nevertheless a rich and unique flora with a unique shrubby vegetation, maquis, and is considered as a biodiversity hotspot. The flora encompasses a large number of metal hyperaccumulating plant species, which are able to accumulate in their leaves metals at concentrations 100 to 1000 times higher than normal plants. We established a plot 20 × 20 m plot on Ferritic Ferrasols where we sampled all individual plants exceeding 1 m height. We measured concentrations of 20 elements on 474 individuals representing 37 species and 22 families. The plot included a large diversity of root symbioses and both Ni and Mn hyperaccumulators. We observed a large diversity of mineral nutrition strategies of plants, and this is in line with the Old Climatically Buffered and Infertile Landscape (OCBIL) theory [1] which predicts nutritional specialization on infertile substrates. Nickel and manganese hyperaccumulation can be two different responses to the same soil conditions. Underground niche partitioning for mineral resources could explain the high alpha diversity observed on adverse geological substrates.

KEYWORDS: *Elementome, hyperaccumulator Ionome, OCBIL, Open ecosystems*

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Functional groups and traits of plants in Mediterranean ultramafic shrublands (Sierra Bermeja, Spain)

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ABSTRACT

Plant functional traits (FTs) are important for understanding plant ecological strategies (e.g., drought avoidance), especially in the nutrient-poor soils of serpentine ecosystems (Díaz et al., 2016). In Mediterranean areas, such ecosystems are characterized by climatic factors (e.g., summer drought) that exert a filtering effect. In our study, we analysed 24 species with varying serpentine affinity, from strictly serpentine plants to generalist plants (Pérez Latorre et al., 2018) from two ultramafic shrublands in southern Spain, considering four FTs: plant height (H), leaf area (LA), specific leaf area (SLA), and stem specific density (SSD). We used principal component analysis to identify combinations of FTs and cluster analysis to define Functional Groups (FGs). We defined eight FGs, which suggests that such Mediterranean serpentine shrublands are composed of species with wide-ranging of FTs. Indicator traits explained 67–72% of the variability based on four strategies: (1) lower H than in other Mediterranean ecosystems; (2) middling SSD; (3) low LA; and (4) low SLA due to thick and/or dense leaves, which contribute to long leaf survival, nutrient retention, and protection from desiccation and herbivory. Generalist plants had higher SLA than obligate serpentine plants, whereas the strictly serpentine plants showed more drought avoidance mechanisms than the generalists. The strictly serpentine plants in the serpentine shrublands studied could present greater resilience to climate change, particularly to severe drought, given the presence of their drought avoidance mechanisms compared with generalists, and the high number of FGs identified. Studies of FT, and specially FG, may be very useful for the joint management of species in serpentine ecosystems.

KEYWORDS: *ultramafic vegetation, functional traits, drought avoidance*

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Parallel adaptation to ultramafic soils in *Solidago virgaurea* L. across the Japanese Archipelago

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ABSTRACT

Parallel evolution is an evolutionary consequence of similar adaptations repeatedly arising in different lineages under natural selection. Understanding the processes and mechanisms of parallel evolution can provide significant insight into biological evolution. Soil is a major factor that affects plant survival and fitness, and can exert disruptive selection among different soil types. In this study, we focus on the history of plant microevolution in disjunct ultramafic soil areas of the Japanese Archipelago. The archipelago harbors numerous ultramafic soil patches scattered over 1,500 km. *Solidago virgaurea* L. (Eurasian goldenrod) is a common herbaceous plant growing on both non-ultramafic and ultramafic soils. Ultramafic ecotypes have unique characteristics in their morphological traits, their tolerance to toxic metal ions, and their phenology, which are divergent from non-ultramafic populations. Ultramafic populations show similar phenotypes even in geologically distant regions, yet phylogenetic origin of these populations remains unknown. To determine whether the ultramafic ecotypes of *S. virgaurea* have single or multiple origins, we conducted genetic analyses of paired non-ultramafic and ultramafic ecotype populations from eight regions in the archipelago. Phylogenetic analysis showed that the ultramafic ecotypes were polyphyletic and closely related to the paired non-ultramafic populations in each focal region. Genetic differentiation was low between adjacent non-ultramafic and ultramafic populations, with an average of $F_{ST} = 0.058$ (range: 0.033-0.095). These results suggest that ultramafic ecotypes are derived from ancestral non-ultramafic types in every region via parallel evolution, and that unique phenotypes of ultramafic ecotypes have been maintained despite gene flow from surrounding non-ultramafic populations. These paired soil ecotypes will be suitable biological replicates to investigate specific genomic regions evolving under disruptive selection during parallel soil adaptation.

KEYWORDS: parallel evolution, ultramafic soil, natural selection, multiple origin, soil adaptation

The elemental defence hypothesis: Is there a ‘missing piece’?

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ABSTRACT

Metal(loid) hyperaccumulation may be one of the most extreme traits as a result of plant adaptation and evolution towards exceptionally high concentrations of metal(loid)s in soil^[1]. A long-standing *raison d'être* for metal(loid) hyperaccumulation is considered in the elemental defence hypothesis (EDH), which postulates that metal(loid) hyperaccumulation exerts an effective inorganic defence against various antagonistic attacks^[2]. Just like many other plant species, hyperaccumulators also produce a broad range of specialised metabolites acting as organic defences and these can greatly vary in the concentration and composition within species and even within an individual, termed chemodiversity^[3]. Such considerations of chemodiversity, however, are currently still missing in the EDH. Thus, we aim to integrate this ‘missing piece’ through the lens of plant chemodiversity to better understand the eco-evolutionary dynamics and maintenance of metal(loid) hyperaccumulation.

We collated and examined relevant research papers about metal hyperaccumulation in the context of EDH over the past 42 years. We found that: (1) inorganic and organic defences are highly diverse; (2) they are metabolically intertwined, as particularly well studied in Brassicales hyperaccumulators; (3) trade-off or joint effects between inorganic and organic defences can occur, but without consistent patterns across studies; and (4) both types of defences can be induced upon antagonistic attacks. Moreover, we revealed that edaphic, population, ecotypic, temporal and spatial related factors apparently influence the elemental defence diversity of hyperaccumulator plants, which again, are often neglected in the EDH. Our findings imply that the EDH should incorporate these various aspects of chemodiversity. Finally, we synthesise a novel framework and outlook to better understand the many fascinating aspects of elemental defence by extending it in the light of plant chemodiversity. With this novel synthesis, we hope to stimulate progressive research and discussions about the EDH and whether this can be a useful consideration for phytoremediation efforts.

KEYWORDS:

chemodiversity; elemental defence hypothesis; hyperaccumulator; inorganic defence; metal(loid) hyperaccumulation; organic defence; specialised metabolite

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The role of metal accumulation in serpentine adaptation in the group *Alyssum montanum s.l.*

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ABSTRACT

Serpentine soil is one of the strongest challenges in nature for plants. It is naturally toxic, thanks to a high concentration of heavy metals, propensity to drought, and shortage of nutrients. In order to colonize serpentine soil, plants often have to evolve and (locally) adapt through special molecular and physiological traits. One such adaptation may be hyperaccumulation: the ability of a plant species to take up large amounts of heavy metals and storing them in its tissues. This ability is of particular interest for human purposes, as, if exploited, it can help restore damaged ecosystems through phytoremediation. Species encompassing populations inhabiting both non-toxic as well as toxic soils might provide ideal study systems for exploring the evolution of hyperaccumulation and adaptation towards toxic soils. In general, hyperaccumulators are mostly found in the Brassicaceae family and especially in the genus *Alyssum s.l.* We here tested two understudied species, *Alyssum gmelinii* and *Alyssum spruneri*, representatives of the typical hyperaccumulator tribe *Alyssae* (Brassicaceae), as promising new model species to untangle the role of metal (hyper)accumulation in adaptation to serpentine soil and establish their unknown phytoremediation potential. The species are distributed along Central Europe and the Balkans in both serpentine and non-serpentine environments. We performed a transplant experiment and tested the differences in fitness between the populations and metal accumulation in the plants' tissues. Moreover, we grew the same populations in anthropogenically polluted soil to assess the phytoremediation ability of the species. Our preliminary results show fitness advantages of plants originating from both types of soil in their substrate of origin. A subset of populations analyzed for Ni uptake shows *A. gmelinii* is likely not a hyperaccumulator, although there is considerable natural variation in the ability to accumulate Ni among populations.

KEYWORDS: *adaptation, hyperaccumulation, phytoremediation*

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Ultramafic ecosystems as a macroecological model

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ABSTRACT

Ultramafic ecosystems are renowned for high endemism and habitat specialization. Although ultramafic ecosystems are important model systems for ecological and evolutionary theory, their importance in macroecology has not yet been fully realized. We argue that ultramafic ecosystems create an appealing and tractable macrosystem for understanding global patterns of plant diversity and responses to land use and climate change. We provide an overview of how such a global macrosystem could be used to understand interactions between climatic and edaphic properties and their influence on plant form and function at a global scale. Furthermore, we propose new avenues of interdisciplinary research that emerge from a macro-scale approach and that create new opportunities for international collaborations to understand these diverse ecosystems. Specifically, we provide three examples of this approach: first, by highlighting a recent synthesis of the edaphic control on plant diversity (Hulshof and Spasojevic 2020); second, by demonstrating possible soil-climate interactions and the diverse plant form and function of tropical ultramafic ecosystems (Garnica-Díaz et al. 2022); and lastly, by summarizing a comparison of a key plant functional trait – specific leaf area – across different biogeographic regions (Samojedney et al. 2022). We end with a ‘call to action’ to compile and disseminate a global GIS database of ultramafic soils which will enable future collaborations to address these types of macroecological questions.

KEYWORDS: *climate-soil interactions; global; macroecology; plant ecology; synthesis*

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Above and belowground functional space of herbaceous serpenticolous species

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ABSTRACT

Serpentine ecosystems are described as stressful because plants face multiple constraints, including lack of nutrients, shallow soils, and trace element toxicity. Therefore, functional ecology theory predicts that serpenticolous species should exhibit attributes similar to those of species found in other harsh environments, namely short size (plant stature functional axis) and slow soil-resource acquisition corresponding to conservative species (leaf economic spectrum axis)^{1,2}. Our main objective was to verify if herbaceous serpenticolous species found in western Europe were short and conservative as expected. Additionally, we investigated acquisitive root traits related to mycorrhizal association and root growth^{3,4}. We sampled above- and below-ground traits of 46 species in 6 serpenticolous herbaceous communities in the French Massif Central and the Apennines (Italy) in 2022. Aboveground traits were positioned in the global functional space provided by a large database of aboveground plant traits. Principal component analysis on all measured traits provided additional insights regarding functional variation of studied species. If serpenticolous species had small size (as expected), they showed important variation regarding leaf economic spectrum and were not particularly conservative compared to herbaceous species sampled worldwide. This raises the question of how corresponding species acquire nutrients in such harsh environments. Root trait analysis showed a large variety of strategies, with species from the Apennines relying more on mycorrhizal association and species from the Massif Central having faster root growth. This might be due to the biogeographical context and, in particular, the occurrence of summer drought in the Mediterranean climate of the lower Apennines. Future work should investigate the correspondence of these functional variations with another tenet of functional adaptation of serpenticolous species: leaf metal accumulation or exclusion.

KEYWORDS: *Plant form and function; Leaf Economic Spectrum; Root functional space; Serpenticolous species.*

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Genetic parallelism during recurrent serpentine adaptation in the Eurasian goldenrod in Japan

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ABSTRACT

Serpentine soils are characterized by an imbalance of soil elements and excess of toxic metals that impose obstacles for successful colonization of plants. Therefore, adaptation to the extreme soil is a fundamental process in the evolution of serpentine plant species. During evolution of serpentine and non-serpentine populations, non-random mating can lead to the establishment of reproductive isolation, which sometimes results in repeated generation of serpentine adapted populations on different isolated islands of serpentine substrates. When such parallel evolution occurs in relatively short evolutionary terms, genetic variations pre-existing in ancestral populations are expected to be reused by natural selection. Yet, our knowledge is limited regarding how standing variation is selected during serpentine adaptation, in which other processes such as gene flow, drift, and dispersal limitation are also involved. The Eurasian goldenrod (*Solidago virgaurea* L. s.l.) is widespread in Japan, and serpentine populations showing edaphic tolerance and early flowering habit repeatedly evolved in isolated serpentine areas. Targeting the serpentine *Solidago* populations and their surrounding non-serpentine counterparts, we first aimed to identify candidate genes underlying serpentine adaptation by whole genome sequencing of two ecotypic population pairs. The allelic variations at these genes were then determined for paired serpentine and non-serpentine populations in 12 locations to investigate the extent of genetic parallelism over the regional landscape. As the results of genome-wide association analysis of SNPs and soil types, 12 genomic regions scattering at nine pseudochromosomes showed significant levels of association. These regions included multiple genes encoding ion transporters (e.g. K, Ca, Mg transporters) and phospholipases, indicating the importance of ion homeostasis and signal transduction during serpentine adaptation. Genotyping at eight candidate genes revealed that derived alleles were consistently more dominant in serpentine populations. Genetic parallelism across locations was most pronounced for one Mg transporter gene. Notably, the allelic distributions at these candidate genes were in sharp contrast to the spatial genetic structure inferred from neutral markers, which only showed a pattern of isolation by distance. Overall, our genomic assessment suggested that serpentine adaptation has a polygenic basis in the *Solidago* populations and, therefore, the convergent genotypic constitution at the adaptive genes was likely shaped by recurrent natural selection in spite of imperfect genetic linkage and on-going gene flow.

KEYWORDS: *Ion transporters; Parallel adaptation; Solidago; Whole genome sequencing*

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Tolerance to ultramafic soils is mediated by control of Ca:Mg ratio in *Santolina semidentata* and associated taxa

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ABSTRACT

Santolina semidentata is a shrub native to the NW Iberian Peninsula which is able to grow on different types of substrates including ultramafic areas. *Santolina melidensis* is a serpentinite-endemic closely related species, and both taxa are members of the *Santolina rosmarinifolia* complex [1]. In order to study tolerance to ultramafic soils in *S. semidentata*, we conducted a reciprocal transplant experiment in which seeds from three populations of *S. semidentata* (from three different soil types), one population of *S. rosmarinifolia*, and the unique population of *S. melidensis* were germinated and grown for 18 weeks in three different soils: one derived from limestones, one derived from granodiorites, and one derived from serpentinites. At harvest, we measured shoot and root biomass and quantified foliar concentrations of Al, Ca, Co, Cr, Fe, K, Mg, Mn, Na, Ni, P and Zn. Effects of the experimental factors soil, population, and the interaction soil * population, on plant biomass were assessed by a split-plot design fitted by a mixed linear model, whereas a principal component analysis (PCA) was used to explore differences in leaf ionomes. Plant survival on limestone and granodiorite-derived soils was around 100% for all five populations. In contrast, the serpentine endemic *S. melidensis* and the ultramafic population of *S. semidentata* were the only plants able to survive and thrive on the serpentinite-derived soil. There were significant differences in plant biomass between populations, with the serpentine-endemic *S. melidensis* having lower biomass. Soil type had a major effect on leaf ionomes of the five populations. However, the two ultramafic populations showed consistently higher Ca concentrations and lower Mg concentrations than the three non-ultramafic populations. This was particularly marked in plants growing on the limestone-derived soil, where ultramafic populations had a Ca:Mg foliar ratio around 25 vs. values lower than 8 for non-ultramafic populations. Moreover, the ultramafic-endemic *S. melidensis* showed unusually high sodium foliar concentrations (around 1% in plants from the three soils). Our results suggest that colonisation of ultramafic soils has involved the evolution of a tolerant ecotype in *S. semidentata*. This tolerance seems to be mediated by efficient mechanisms of regulation of the balance between Ca and Mg. High accumulation of sodium in the leaves of the ultramafic-endemic *S. melidensis* may be related to mechanisms of tolerance to drought stress in ultramafic areas.

KEYWORDS: *Santolina rosmarinifolia* complex, reciprocal transplant experiment, calcium:magnesium ratio

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Lichen diversity and abundance on serpentinite and granite outcrops of the Slavkov Forest (Czech Republic)

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ABSTRACT

This study is part of the eLTER PLUS project DiESCALiDi (Disentangling the Effects of Substrate Chemistry and Atmospheric Deposition on Lichen Diversity), which aims to evaluate if and how saxicolous lichen communities may be informative on pollution scenarios. Saxicolous lichens were indeed recognized to suffer chemical depositions similarly to epiphytic lichens [1]. However, higher system complexity related to the additional influence of the rock substrate chemistry made them generally disregarded as potential biomonitors of the effects of air pollution, although in several circumstances they may be a valid or the sole alternative to obtain data. In order to (re-)consider potential relationships between the diversity and abundance of saxicolous lichens and different conditions of air pollution, the first step is thus to evaluate how the nature of the rock substrate and other environmental conditions could potentially affect lichen colonization. In this work, we compared patterns of lichen diversity and abundance on serpentinite and granite outcrops inside and outside forest stands in the Lysina and Pluhův Bor catchments and some adjacent areas of the Slavkov Forest (W-Czech Republic), sharing meso-climate conditions and atmospheric deposition fluxes. The frequency and cover of lichen species were surveyed through 42 plots of 50×50 cm², distributed in nine sites representative of the mentioned conditions, using a square grid divided into 25 quadrats. At each site, the diversity of epiphytic lichens was also surveyed, and thalli of the saxicolous species *Parmelia saxatilis* (L.) Ach. and the epiphytic species *Hypogymnia physodes* (L.) Nyl. and *Hypogymnia tubulosa* (Schaer.) Hav. were collected for chemical analyses. GLM models showed that both substrate lithology and the presence/absence of the canopy significantly affect the saxicolous lichen cover, while only the latter factor is significant for the lichen alpha-diversity. In particular, a higher number of saxicolous species characterized plots outside the forest stands, with similar values for granite and serpentinite. Consistently, also cover values both on granite and serpentinite outcrops were higher outside forest stands. These preliminary findings suggest that, at least in the study area, substrate lithology is not a driver of saxicolous lichen richness. On the other hand, a canopy cover, and hence presumably light influx, is very important for saxicolous lichen diversity. The relation between different lichens species' functional traits, including secondary metabolites, and serpentine features will be discussed to better understand their adaptation, as well as the presence of nitrophilous species as a remarkable component of saxicolous lichen communities outside forest stands.

KEYWORDS: *saxicolous lichens, air pollution, biomonitoring, functional traits, lichen adaptation, serpentinites vs. granites*

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Community assembly of serpentine plants is driven by interspecific differences in functional traits.

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ABSTRACT

Serpentine ecosystems are characterized by multiple environmental stressors such as high levels of heavy and trace metals (e.g. Ni, Cr, Co and Mg), as well as low availability of macronutrients (e.g. N, P and K) and low water retention. This high contents in toxic metals exert a strong filtering effect on plant communities, selecting species and traits adapted to these harsh conditions. To better understand the effect of stressful environmental conditions on plant strategy and community assembly processes, we studied 26 plots in serpentine (S) and non-serpentine (NS) sites constituting a metal gradient, and we focused on the dominant plant species. We measured 6 leaf functional traits related to resource acquisition and stress resistance on 20-100 individuals: leaf area, specific leaf area, leaf thickness, leaf dry matter content, leaf nitrogen content and leaf shape. We quantified the proportion of variance explained by the interspecific traits difference, intraspecific trait variation explained by levels of soil metal, and residual variation. We tested if individual species show differences in trait values between high and low metal soils. At the community level, we calculated the weighted mean value of each trait to test if the community is dominated by stress tolerant strategies (small thick leaves with low SLA and N, high dry matter content and large length/width ratio). Using several uni- and multivariate functional diversity indices, we tested if we could detect some trait convergence on soils with high levels of metals, suggesting environmental filtering, and traits overdispersion on low-metal soils which could indicate niche partitioning. Finally, we ran all the analyses on the same dataset after excluding the only serpentine endemic and metal hyperaccumulating species, *Odontarrhena lesbiaca*. For leaf area, thickness, dry matter content and shape most of the variance is explained by interspecific trait differences, while for specific leaf area and nitrogen content, a significant proportion of the variance is explained by intraspecific variation due to soil metal levels. However, most species do not show differences in trait values between high and low metal soils (only 1 to 3 species per trait). At the community level, all weighted mean trait values exhibit differences between soil types, with convergence to a stress tolerance syndrome (small leaves, low SLA, thick leaves with high LDMC), although LNC was surprisingly higher on high metal soils. Regarding multivariate FD indices, contrary to our hypothesis, only sesFD_{is} shows a significant higher value on high metal soils. Using univariate sesFD_{is} index, LDMC and LS shows the expected convergence, but SLA and LT show clear overdispersion on high metal soils. When removing *O. lesbiaca* from the studied community, both multivariate sesFR_{ic} and sesFD_{is} showed the expected convergence on high metal soils soils. With univariate sesFD_{is}, SLA does not show any clear pattern of convergence or divergence, while LDMC and leaf thickness shows some level of convergence. Our results suggest that high metal soils do not drive strong intraspecific variation for traits related to the LES for most species. Further, a single endemic species with original traits can drive the whole community level indices of functional diversity. We suggest caution in the interpretation of these indices in the context of harsh environments where plant adaptations can be diverse. Further we advocate for a conservation of functional diversity in serpentine environments, notably of endemic species with original traits combination, to ensure resilience in the face of environmental change.

KEYWORDS: *Intraspecific variation, leaf economic spectrum, metal tolerance, environmental filtering, competition*

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Arable plant communities depend largely on soil type: a study case from ultramafic and non-ultramafic soils in Beni Bousera (North Morocco)

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ABSTRACT

Traditional agroecosystems play a vital role in conserving arable plant diversity, which requires the preservation of this ecological heritage. This study aims to compare the species richness, composition, Shannon diversity, and Simpson diversity of arable plant communities in traditional agroecosystems on ultramafic and non-ultramafic substrates in the Beni Bousera massif region of northern Morocco. We surveyed arable plant communities in the study area and conducted a physicochemical analysis of soil composition to explore the relationship between arable plant community composition and soil properties. We observed a significant difference in floristic richness between the two soil types, with a moderate serpentine effect being highly abundant. Among the 167 identified arable plant species, *Asteraceae* and *Poaceae* were the most widespread botanical families. Therophytes were the dominant life forms at both study sites, followed by hemicryptophytes and geophytes. Non-ultramafic plant communities were generally more species-rich than the ultramafic site vegetation. We found a differentiation in arable plant communities according to substrate type, with a calcifuge-affinity community at the ultramafic site and a calcicole-affinity community at the non-ultramafic site. Our results highlight the differentiation in arable plant communities at the study sites, despite the proximity and similarity of climate and agricultural practices.

KEYWORDS: *Weed community, serpentine effect, the massif of Beni Bousera, traditional agroecosystems, vegetation relevés.*

Disentangling plant ionomes of two pseudo-metallophyte orchid species on contrasting soils in the Balkans

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ABSTRACT

Orchids are known to occur in almost all terrestrial habitats, including on potentially toxic metalliferous soils. *Dactylorhiza sambucina* (L.) Soó and *Gymnadenia conopsea* (L.) R.Br. are widespread terrestrial orchids in Europe, which are generalists, *i.e.*, they inhabit different habitat types on various geological substrates. The high ecological plasticity with respect to soil properties raised the question whether these contrasting geological substrates lead to differences in the elementome profiles and ecophysiological responses of these orchid species. Of particular interest are ultramafic soils characterized by a strong edaphic filter due to their adverse chemical properties. In this context, plants from eight populations of *D. sambucina* and nine populations of *G. conopsea* from three types of substrates (ultramafic, calcareous, and siliceous soils) were analyzed to evaluate differences in ionome, physiological and biochemical parameters. These results are evaluated in light of previous findings suggesting that the uptake of macroelements in aboveground plant organs is predominant, while for trace elements the exclusion strategy is generally found. The analysis of ecophysiological parameters (including concentrations of photosynthetic pigments and phenolic compounds) should clearly reveal any differences between populations from the different geological substrates, with the lowest stress level expected in plants growing on limestone soils. The results of this study are important not only because of the general lack of data on elementome profiles and ecophysiological properties of orchid species on ultramafic substrates, but also from the point of view of the safe use of the underground tubers which are locally used to produce the drink ‘salep’.

KEYWORDS: *Orchidaceae, ultramafic, metal tolerance, physiology*

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Edible plants and aquatic systems in Serpentine region in Bulgaria

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ABSTRACT

The Serpentine soils are formed due to the weathering of ultramafic rock types (peridotites (dunite, wehrlite, harzburgite, lherzolite) and the secondary alteration products formed by their hydration including serpentinite. These soils are generally deficient in plant essential nutrients such as nitrogen, phosphorus, potassium, and sulfur; have a calcium-to-magnesium (Ca:Mg) molar ratio of less than 1; and have elevated levels of toxic elements such as nickel, cobalt, and chromium. Serpentine soils promote the evolution of serpentine endemism, contributing to unique floras with high rates of endemism. Ecosystems with serpentine soils are generally less productive than ecosystems with other kinds of soils, and they have unique plant species distributions. However these type of soils might be also used for edible plants growing although with less productivity and the question arising is – what will be the content of essential and toxic content in the plants as well as what will be the influence on the local aquatic systems frequently used for weathering of agriculture field. In the present study the concentration of chemical elements ((Ca, Mg, Fe, Mn, Zn, Cr, Cd, Pb, As) was determined in aqueous phase and biota in rivers from serpentine region in Bulgaria, situated also in the vicinity of abandon chromium mines. The content of essential and toxic elements (Ca, Mg, Fe, Mn, Zn, Cr, Cd, Pb, As) was determined in edible plants (*P. vulgaris*, *Cucumis sativa*, *Capsicum annum*, *N. tabacum*, *Lycopersicon esculentum*, *Solanum tuberosum*) grown in the same region. Samples are collected in one season, chemical elements in river water was determined by ICP-MS, chemical elements content in plant, biota was determined after MW digestion and ICP-OES measurement. Results obtained will be discussed from the view point:

- Correlation between bioavailable concentration of chemical elements in soil and their content in edible plants.
- Correlation between bioavailable concentration of chemical elements in soil and in aquatic systems – aqueous phase used for plant weathering and biota.

Conclusions will be directed to hazard assessment of toxic elements content in edible plants and in biota used for human consumption.

KEYWORDS: *serpentine soil, chemical element, river water, biota*

Acknowledgment: Financial support from project EXTREME, funded by the Bulgarian Ministry of Education and Science (DOI-76/30.03.2021) is highly acknowledged.

Exploring the drought tolerance hypothesis of nickel hyperaccumulation using *Berkheya* species of South Africa

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ABSTRACT

Serpentine soils have extreme physical and chemical properties which generally create a hostile environment for normal plant growth. Species growing in these soils are adapted to deal with high levels of bioavailable metals, including nickel. To date, over 500 metallophytes are known to hyperaccumulate nickel, including a total of six species from South Africa (*Berkheya coddii*, *B. zeyheri*, *B. nivea*, *Senecio anomalo-chrous*, *S. coronatus* and *S. conrathii*). Metal hyperaccumulation is an energy costly process for plants. It is therefore expected that this trait has an adaptive or intrinsic fitness benefit. Several major hypotheses have been proposed to explain the adaptive significance of this trait, including elemental defence, inadvertent uptake, nutritional benefit, elemental allelopathy, and drought tolerance. In serpentine tolerant plants, there is significant selective pressure for drought tolerance. It is hypothesized that nickel ions may act as an osmoticum in hyperaccumulators, where they aid in adjusting the plant osmotic potential under water stress. We aim to test the drought tolerance hypothesis using one Ni-hyperaccumulating taxon from South Africa (*B. coddii*) and a native congener that is tolerant of serpentine soil, but is not a hyperaccumulator (*B. radyeri*). The experiment will be done in a greenhouse where these taxa will be exposed to high, medium, and low levels of water stress in serpentine soil and potting soil with and without nickel added. Photosynthesis/transpiration rates and leaf nickel content will be measured at key phenological stages of the plants (three month old seedlings, pre-flowering, fruiting, and senescence) while biomass (root and shoot) and total reproductive output (total number of flowerheads) will be measured at the end of the experiment. We hypothesize that the plants growing in nickel-enriched soil (serpentine and potting soil with nickel) will show greater carbon acquisition and growth and reproductive fitness under high water stress compared to those in the potting soil without nickel. While we believe the nickel hyperaccumulating taxon will benefit the most under drought, the serpentine-tolerant taxon may also show better growth and fitness under water stress in the presence of nickel. This study will contribute to our understanding of the little-explored drought tolerant hypothesis of metal hyperaccumulation, and shed light on evolutionary consequences of edaphic specialization. Additionally, this study can also contribute to restoration efforts, including phytomining and phytoremediation.

KEYWORDS:

adaptation, geoecology, heavy metals, metal tolerance, phytoremediation, Senecio

The effect of substrate type and species richness on biomass production and nickel concentration

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ABSTRACT

Biodiversity is considered to be able to mitigate the detrimental effects of global change drivers on ecosystem functioning and this beneficial impact has been found to increase with increasing biotope space [1]. Biotope space can be defined as the physical space in which species niches may accommodate (e.g. depth or volume of soil), or in the case of resource availability it can have a chemical, spatial or temporal dimension (e.g. availability of different types of nutrients, from different soil layers, at different times). A reduction in biotope space under conditions of intense physical stress may reduce the positive biodiversity effects [1, 2]. Serpentine soils are considered stressful environments for plant growth [3]. In this study, we investigated the interaction of biotope space and species richness on aboveground biomass and the nickel concentration. Three substrates of increasing stress intensity and decreasing biotope space were used: peat, mixture of peat and serpentine soil and pure serpentine soil. In each of these, monocultures and bicultures of the endemic plant species *Odontarrhena lesbiaca* (perennial species, nickel hyperaccumulator) and *Alyssum xiphocarpum* (annual species, non-hyperaccumulator) were grown in pots for one growing season. At the end of the growing season, aboveground biomass and nickel concentration in plant organs as well as total and bioavailable soil nickel concentration were measured. Peat had the highest percentage of bioavailable nickel, while having much lower total nickel concentration than the mixed and serpentine substrates. Total biomass per species and per pot was influenced by the total number of individuals survived, species diversity and substrate type. Substrate type and species identity significantly affected the nickel concentration in plant tissues, while diversity was statistically insignificant. No significant interaction between diversity and soil substrate type was observed in both biomass and nickel concentration. Substrate type seems to have the greatest effect on the measured variables, with peat being the best substrate for plant growth.

KEYWORDS: *biotope space, pot experiment, species diversity*

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Ultramafic lichens: an ecological investigation of an understudied organism in a well-studied system.

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ABSTRACT

Saxicolous lichens are a major, ecologically significant component of the biodiversity present in rock outcrop habitats, including ultramafic outcrops. While multiple studies have investigated the biotas of ultramafic outcrops, few have directly compared ultramafic and non-ultramafic lichen communities^{1,2,3}. As a result, the importance of substrate factors (elemental composition, surface pH, microtopography) in relation to other abiotic factors is poorly understood for saxicolous lichen communities. In a regional community ecology study, we sampled lichen biotas of eight ultramafic and eight sandstone outcrops along a 70 km maritime influence gradient in order to test three hypotheses: 1) a *substrate effect hypothesis* that saxicolous lichen communities of ultramafic and sandstone outcrops are compositionally distinct; 2) a *maritime gradient hypothesis* that coastal and inland communities are compositionally distinct; and 3) a *maritime moderation hypothesis* that coastal ultramafic and sandstone communities are more similar than those of inland ultramafic and sandstone.

Our results provide support for the *substrate effect hypothesis* and the *maritime gradient hypothesis*. Lichen communities of 1) ultramafic and sandstone outcrops and 2) coastal and inland outcrops were compositionally distinct. The *maritime moderation hypothesis* was not supported; lichen communities of ultramafic and sandstone outcrops were significantly differentiated throughout the maritime gradient. A total of 134 taxa were recorded - 81 taxa from ultramafic outcrops and 100 taxa from sandstone, with 47 taxa found on both rock types. Ultramafic outcrops were characterized by greater similarity between samples, lower lichen cover, larger differences in cover between north and south aspects, and higher abundance and diversity of cyanolichen taxa compared to sandstone. This study is one of few to quantitatively examine lichen communities of two rock types and is unique in that it does so at a regional scale. These results add to our understanding of the interactive roles of substrate and maritime influence in lichen community assembly.

KEYWORDS:

lichen, substrate effects, maritime gradient

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A microscopic description of the actinorhizal nodules of Casuarinaceae trees growing on ultramafic areas in Sabah (Malaysia)

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ABSTRACT

Casuarinaceae (Order Fagales) is a plant family distributed in the Malesian-Australian-Melanesian region and comprises four genera (ranked from more basal to more recent): *Gymnostoma*, *Ceuthostoma*, *Casuarina* and *Allocasuarina* [1]. At the landscape scale, Casuarinaceae act as pioneer trees on degraded or nutrient-poor soils including mine tailings and ultramafic areas. This pioneer nature is possible thanks to different traits related to nutrient conservation (e.g. extreme leaf reduction, presence of clusteroid roots) but also to the presence of actinorhizal symbioses with *Frankia* N₂-fixing actinobacteria. The Casuarinaceae-*Frankia* symbiosis is an intriguing model of evolution of plant-microbe interactions. Different studies have confirmed that *Gymnostoma* established a nonspecific symbiotic association with easy-to-isolate, more saprophytic *Frankia* strains which belong to Cluster 3 frankiae, whereas species of the genera *Casuarina* and *Allocasuarina* are in symbiosis with hard-to-isolate and slow-growing *Frankia* strains from the Cluster 1c frankiae [2]. Moreover, the *Frankia* in *Gymnostoma* root nodules produce microscopical thick-walled vesicles where nitrogenase is protected from oxygen and nitrogen fixation occurs, whereas the *Frankia* in *Allocasuarina* and *Casuarina* root nodules are able to fix nitrogen without need to produce vesicles thanks to special structures developed by the plant host and to the synthesis of hemoglobin. In contrast, the presence of actinorhizal nodules in *Ceuthostoma* (a genus which is in an intermediate position between the other three Casuarinaceae genera) has been only confirmed recently [3] and no information exists on the anatomy or the type of *Frankia* strain associated to this plant genus.

In order to obtain first evidences on the *Frankia* strains associated to the genus *Ceuthostoma*, actinorhizal nodules were collected on the roots of several plants of *C. terminale*, *Gymnostoma nobile* and *G. sumatranum* from ultramafic areas in Kinabalu Park (Sabah, Malaysia). Nodules from *Casuarina equisetifolia*, which grows on sandy beaches in Sabah were also collected for comparison. Semi-thin sections of nodule lobes were obtained, stained with lactophenol blue and observed under the microscope to determine i) the distribution of *Frankia* infected cells along the lobe and ii) the presence or absence of bacterial vesicles.

The obtained results will be discussed within the framework of the evolution of actinorhizal symbiosis in the whole family Casuarinaceae.

KEYWORDS:

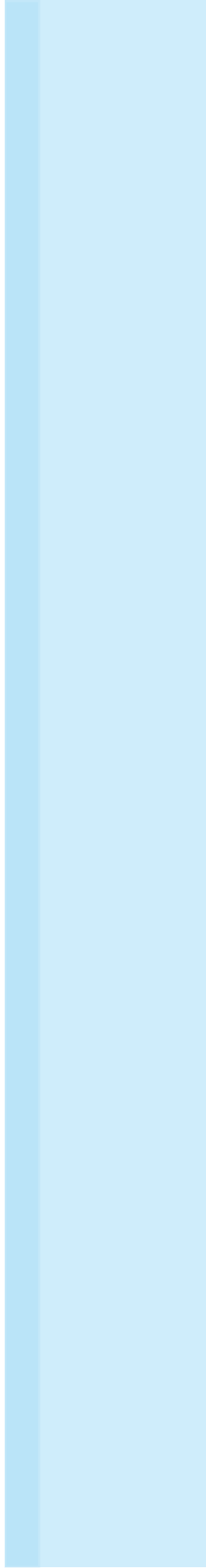
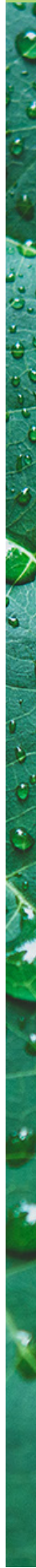
Casuarinaceae, *Frankia*, root nodules, semi-thin section, symbiosis

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Session 4 - Conservation & restoration



Revegetation of mine tailings in the abandoned asbestos mine of Balangero and Corio (Torino, Italy): Twenty years after

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ABSTRACT

The largest chrysotile asbestos mine in Western Europe, located in the Ultramafic Lanzo Massif (internal Western Alps, Italy), stopped its activity in 1990. Nevertheless, the mining area (approx. 400 ha), and particularly the open pit mine and million tons of mine tailings, remained a source of airborne asbestos fibres. In 1998, the former asbestos mine was recognized as a 'contaminated site of national interest' and remediation activities started. Interventions on the main volumes of mine tailings, dumped on the southern (SEMTs) and northern (NEMTs) side of the mountain area, were conducted in 2000-2003 and 2004-2009, respectively, and included hydro-geological stabilization and revegetation practices. In particular, hydroseeding of non-serpentine herbs was associated with the planting of shrubs and trees. A non-serpentine topsoil and mulch were added on SEMTs, while hydroseeding was performed directly on the serpentine debris of NEMTs. A vegetation survey conducted in 2003-2004 already evaluated dynamic stages of spontaneous colonization in different areas of the mine [1]. However, the SEMTs had been just reprofiled and were still bare; the NEMTs still needed reprofiling and the high steepness and consequent instability prevented any colonization. In this new study, plant diversity and abundance were surveyed on the SEMTs and NEMTs approximately 20 and 15 years after the respective remediation interventions (n=13 plots of 5×5 m²). The survey also re-considered the horizontal terraces of the open pit mine (n=3 plots), which in 2003-2004 displayed plant communities poor in species and cover, and semi-natural areas, never directly impacted by excavation or discharge activities. Image analysis by color-based pixel classification was used to quantify total plant cover. The SEMTs hosted the highest specific diversity (64 species; av. 16 per plot), similar to that observed in the semi-natural areas, while both the NEMTs (27 species; av. 11 per plot) and the open pit (15 species, av. 8 per plot) displayed lower values. Classification and ordination analyses displayed a higher relationship of NEMTs with the semi-natural area, and of the SEMTs with the open pit mine. Plant cover was significantly higher on the NEMTs (av. 60%) with respect to the SEMTs (av. 35%), while the open mine pit maintained the same low values quantified in the previous study (av. <10%). Environmental factors associated to the northern exposure, likely dealing with water availability, resulted in the primary drivers of plant colonization, rather than the time since intervention. Remarkably, only 30% and 13% of species in the adopted hydroseeding mixtures were recovered on SEMTs and NEMTs, which contributed 10% and 1% of plant cover, respectively. Instead, particularly on the NEMTs, species dominating the semi-natural serpentine grassland in the surroundings (mostly *Koeleria vallesiana*, bryophytes and terricolous lichens) were responsible for most of the cover. In conclusion, chrysotile bearing tailings have good vegetation cover after 20 years (mainly for NEMT), which together with the addition of topsoil (SEMT), account for a drastic decrease of airborne fibres in the mine area. Different patterns observed on NEMTs and SEMTs will be discussed to address potential improvements in revegetation strategies.

KEYWORDS: *chrysotile asbestos mine; fibre dispersion; hydroseeding; mine tailings; plant cover; revegetation*

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Characterising soil microbial diversity for conservation and restoration using large-scale DNA-based methods in New Caledonian ultramafic ecosystems

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ABSTRACT

Owing to their crucial role in ecosystems functioning and their ability to rapidly respond to environmental changes, soil microorganisms could be one of the principal actors for conservation and ecological restoration. However, this hidden biodiversity has been, to date, widely neglected, particularly in the southern hemisphere. New Caledonia is an archipelago located in the southwest Pacific renowned for its exceptional biodiversity, and recognized as a world priority area for conservation and restoration. With one third of its territory covered by soils originated from ultramafic rocks, which contain 20-30% of the global nickel ore reserve, mining has occurred for more than a century and has drastically increased in the past decades. Mining has led to massive land degradation throughout the entire main island. In this context, we conducted several studies on soil fungal and bacterial communities within diverse ultramafic ecosystems, with the aims to (i) characterize the microbial diversity at different geographical scales and taxonomic levels, (ii) identify the main biotic and abiotic factors influencing soil microbial communities, (iii) determine their functional roles, and (iv) infer their potential use as tools to monitor restoration progress and success. Our research relied on different molecular approaches, especially the emergent and revolutionary molecular methods of next-generation sequencing technologies (NGS). Our results revealed the existence of a remarkable soil microbial diversity, notably of the symbiotic group of ectomycorrhizal fungi, which may harbour an endemism rate of 87-95%. Plant and site effects on both fungal and bacterial communities were also highlighted. The site effect suggests that each location, or ultramafic massif, displays its own soil microbial community, and could thus represent distinct conservation units. Microbial composition has also been shown to influence ultramafic soils' susceptibility to erosion. Our recent findings pointed out the major interest of investigating soil microbial communities via the use of high-throughput amplicon sequencing of environmental DNA (eDNA) to monitor ecosystems recovery. We also showed that soil eDNA metabarcoding could contribute to the identification of suitable symbionts for plant inoculation. Overall, our research works highlight the importance of considering soil microorganisms and the emergent new sequencing technologies in conservation and restoration.

KEYWORDS: Biodiversity; eDNA metabarcoding; high-throughput sequencing; monitoring; New Caledonia; soil microbial communities.

Even edaphic specialists can die from climate extremes: Plant mortality on ultramafic soils after a heatwave and intense drought

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ABSTRACT

The frequency, intensity and duration of heatwaves and drought spells are increasing at the global scale, and climate change models predict a further increase of these events in the second half of this century. Plant mortality is increasingly reported from many parts of the world, but no data exist for non-forest communities of edaphic specialists such as those of ultramafic soils. These soils are subject to unusual extremes of temperature, caused by the radiant losses and gains during the diurnal cycle consequent to their typical dark color and large amount of bare ground. Their porous, granular texture permits elevated aeration and causes a strong microclimatic effect, which can be amplified during periods of anomalous heat and drought. This also causes a strong drainage and moisture deficiency. Though serpentine plants are usually well adapted to these conditions, their tolerance cannot be unlimited and one may wonder how they respond when intensity of these stress factors increases during heatwaves and drought spells. Accordingly, we investigated the impact of these events on serpentine vegetation [1]. We assessed mortality of perennial plants at the end of the extreme event of 2017 in ten ultramafic areas along a SW-to-NE gradient in Central Italy. Mortality was unexpectedly high and increased with decreasing number of rainy days and duration of the heatwave. It decreased with increasing distance from the Tyrrhenian coast, and was highest in areas with Mediterranean climate, where mortality reached 60% of total sampled individuals. Plant survivorship was higher on deeper soil and north-facing slopes. Species responses were independent from taxonomic position and functional type. Obligate serpentine endemics were not less impacted than non-endemics. Mortality decreased with increasing species chromosome number, suggesting higher tolerance in taxa of polyploid origin. In 2020, we recorded soil and air temperatures in three sites along the same gradient, which revealed a strong microclimate effect experienced by serpentine plants. Overall, results suggest that heat and drought events can become a threat to species survival, maintenance of population size, and, in the long term, diversity and composition of ultramafic plant communities.

KEYWORDS: *Drought; heat waves; plant mortality; serpentine soil; temperature*

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Implementing Access and Benefit Sharing (ABS) of Genetic Resources (GR) for Agromining in Sabah, Malaysia

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ABSTRACT

Since 2010, a group of international scientists had been studying hyperaccumulative plants which have been growing on barren ultramafic lands in Sabah, Malaysia. After several successful collaborative projects, they have identified several plant candidates to be developed as metal crops. This has triggered the mandatory ABS implementation as stipulated in Sabah Biodiversity Enactment 2000 which is in tandem with the requirement of Nagoya Protocol. The group of researchers had conducted preliminary study under academic research license and the findings indicate that there is good potential to use *Phyllanthus rufuschaneyi* to extract nickel from the earth. Eventually, leading to the formation of Econick Sabah Sdn. Bhd to explore the commercial viability of this agromining. Sabah Biodiversity Centre (SaBC) has drafted out several mutually agreed terms (MAT) in the of ABS agreement which aims at providing access to aforementioned protected plants in addition to supporting research activities which will be carried out by Econick Sabah and local collaborators to utilize the plants in phytoextraction. In return, Econick Sabah will work with local communities by establishing metal farms from which they will receive side income while waiting for their lands to be remediated. Besides non-monetary terms, a few monetary conditions are in negotiation. This paper describes the process of implementing the ABS of *Phyllanthus rufuschaneyi* and a few other hyperaccumulators for agromining in Sabah, Malaysia.

KEYWORDS: *Nagoya Protocol (NP) under Convention of Biological Diversity (CBD), phytoextraction, metal farm*

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Plant communities of Cuban ultramafic soils and the impacts of climate change

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ABSTRACT

Ultramafic communities are highly specialized ecosystems to the substrate that sustains them. Throughout Cuba, these areas are distributed as relatively small and spatially isolated patches representing 7% of the island's surface [1], they could be considered "islands within an island" and harbor a third of Cuban plant endemics. Associated to ultramafic soils there are two types of natural vegetation: scrubs and forests; however, due to human impacts these communities are transformed into savannahs [1]. As result of the climate change, the increase of the annual mean temperature, drought, incidence of fires, and the frequency and intensity of the hurricanes could modify the environmental conditions in these ecosystems [2, 3]. The flora of ultramafic outcrops is mainly composed of facultative (generalist) plants capable of inhabiting ultramafic and non- ultramafic substrates, they therefore have the potential to migrate, but edaphic specialist (endemic) species could be affected by their own limited habitat. Many of these plants have a high degree of xeromorphic traits and a slow growth rate, making them able to adapt to harsh environments. The xerophytes communities as shrubs and pine forests could be more resistant to climate changes because litter and organic matter produce by "stress tolerant plants" is very recalcitrant, decomposes slowly and promotes a slow nutrient cycling. However, climate change could exceed the adaptation capacity of these species, especially since them already occur in abiotically stressful environments [2]. Niche modelling tools have verified that several range restricted plant species of sclerophyllous ultramafic forests in mountain areas should have their areas of climatic suitability reduced in the future [2, 3]. Many of the ultramafic outcrops in Cuba are included in the "Protected Areas System" and the management of these areas could be a key tool to reduce the modifications in the composition and distribution of their flora and communities.

KEYWORDS: *Climatic niche; Protected Areas management; serpentine sclerophyllous forest; specialized plant communities*

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Reproductive life traits of plants growing on ultramafic substrate on the Koniambo massif (New Caledonia) as a tool to reinforce and reconnect fragmented forests.

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ABSTRACT

New Caledonia has the important task to combine its exceptional biodiversity with the rapid development of human activities. One of the most important threats is habitat fragmentation especially due to mining on ultramafic soils [1]. Over the past 40 years, revegetation was the main approach to mitigate such impacts. Ecological restoration capability and concepts have recently been developed in the past 15 years, and requires knowledge of ecological succession and the study of seed ecology for restoration. Following a previous study conducted in the south of New Caledonia [2], we applied a same approach in the north of the territory on the Koniambo massif. The aim was to study reproductive traits (e.g. dispersal syndrome, type of fruit, size and weight of seeds, germination and dormancy types) of species constituting two vegetation types para-(MPF) and pre-forest (PF). Both surround the relics of forest (F) that still existing and which have suffered repeated fires. This suggests that they are stages of ecological succession leading to the humid forest that must be restored. The interest of such an approach is to provide, from the new knowledge acquired, a list of candidate species that can be used for plantings to improve forest edges or to constitute stepping zone corridor. To attain our objectives, we first identified forest zones to be studied on the Koniambo massif. We carried inventories in the forest (F) and in surrounding vegetation (MPF and PF). For all listed species, we characterised 41 of their life traits that led to the completion of our previous database [2]. The analysis of these life traits (with a focus on reproductive ones), crossed with the presence of the species in all vegetation types, led to the selection of 15 candidate species. It was predicted that these species would result in a closed forest canopy and/or attract seed dispersers. Their seeds were harvested and their ecophysiology studied. Knowledge of seed germination techniques allowed for the cultivation of seedlings and were used to for a first plantation of 1 hectare both in MPF and PF. Four species (505 individuals): *Cryptocarya transversa* (Lauraceae), *Ficus austrocaledonica* (Moraceae), *Metrosideros laurifolia* and *Syzygium frutescens* (Myrtaceae) could successfully be cultivated and planted. An analysis of all these data will be presented, together with recommendations, to inform land managers on how to implement ecological restoration projects in a forest environment.

KEYWORDS: *Ecological succession, seed ecophysiology, dispersal syndrome*

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Germination and initial growth characteristics of *Saussurea modesta* (Asteraceae, an endangered serpentine plant in Japan)

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ABSTRACT

The perennial herb *Saussurea modesta* Kitam. (Asteraceae) is an endangered species that is endemic to serpentine fields in western Japan. In this study, we conducted laboratory and culture experiments to study the germination and initial growth characteristics of this species, which is essential information for constructing a conservation strategy for the species. The seeds used for the experiments were collected from a natural population in a serpentine field around Mt. Nekoyama (35°1'45" N, 133°11'45" E), Hiroshima Prefecture, western Japan. Thermal germination characteristics were examined using the convenient screening test system "gradually increasing and decreasing temperature (GT) method" [1]. In the GT method, seeds are subjected to two temperature regimes: a gradually increasing temperature (IT) regime and a gradually decreasing temperature (DT) regime. To assess the effects of storage conditions on germination, we conducted an experiment using two different periods (1 and 4 months) and two moisture conditions (wet and dry-storage). Germination (%) was very low (10%) in seeds stored under wet conditions for one month, whereas seeds stored under wet conditions for a longer period (4 month) showed much higher germination (60%). No germination was observed in dry-stored seeds in any of the experiments. These results indicate that this species has dormant seeds and requires long term cold and moisture stratification to break its dormancy. To examine the initial growth response to light, we conducted culture experiments under different light conditions (relative photosynthetic photon flux density (RPPFD): 100%, 50%, and 10%). Under low light conditions (RPPFD:10%), all seedlings died within three months, suggesting that this species cannot grow in shaded habitats, such as the forest floor. The main habitat of *S. modesta* is grasslands maintained by human activities such as mowing [2]. However, such grasslands have been decreasing in size and have changed to forests because of depopulation in rural areas. This results in a decrease in the habitat of this species. In addition, our experiments indicate that this species requires long wet and cold conditions for germination. The soil surface under the thick snow cover is likely to provide such conditions. The distribution of *S. modesta* is restricted to areas with heavy snow during winter. However, the amount of snow has decreased in recent years because of climate change. Our results suggest that habitats suitable for the growth of this species are decreasing; therefore, urgent conservation measures are needed.

KEYWORDS: *Saussurea modesta*, endangered species, germination, conservation,

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Modeling Species Distributions of Select Obligate Metallophytes in Response to Climate Change

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ABSTRACT

Climate change is threatening ecosystems globally. Species distribution models used to understand the impacts of climate change on plant populations have largely focused on species either of economic importance or with the potential to disperse along a gradient of altered temperature and precipitation. Obligate metallophytes are one group of species that may not be able to disperse in the same way, as the distribution of the soils to which they have specialized will not shift with the climate. Improved understanding of the impact of climate change on these species is needed to minimize the loss of their unique communities. To address this gap, we used MaxEnt species distribution models in R. We referenced a regional database of metallophytes to focus on species which are strict endemics and are categorized as rare, threatened, or endangered. We used current distributions, serpentine soil distribution, elevation maps, and bioclimatic variables as the basis for our models. Preliminary analysis suggests that these species may be lost as a consequence of their narrow niche and the inhospitality of future conditions. This indicates that conservation of metallophytes and similar species may be a more pressing conservation concern than currently recognized.

KEYWORDS: *climate change; metallophyte; endemic; obligate; serpentine; ultramafic; MaxEnt models*

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Phytoremediation and Nurse Potential of Aloe Plants from Ultramafic Areas on Mine Tailings

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ABSTRACT

Mine tailings are a source of potentially toxic metals (PTMs) worldwide. Phytoremediation is a low-cost green technology that uses metal-tolerant plants to extract these contaminants and rehabilitate the soil. In mine tailing restoration efforts, it can be beneficial to introduce species that can facilitate the colonization of other plants (i.e., nurse plant syndrome). In this study, the phytoremediation and nursing potential of two species adapted to ultramafic soil, *Aloe burgersfortensis* and *A. castanea*, were evaluated for the first time. An experiment was performed with aloe plants grown in pots containing potting soil, platinum tailings, and gold tailings. Leaves were assessed for bioaccumulation of PTMs. Seeds of Bermuda grass and African daisy, two successional pioneers, were planted with the aloes and had their developmental parameters evaluated after 30 days. Allelopathic effects were also assessed, with seeds of the pioneer plants infused with root extracts of the aloes from the different soil treatments. *A. castanea* demonstrated greater potential for the bioaccumulation of Cd, Co, Mn, Ni, and Zn in the tailings. The presence of aloes benefited germination rates, leaf count, length, and plant biomass of grasses and daisies in the mine tailings, without significant allelopathic effects. Therefore, aloes—especially *A. castanea*—should be employed in the rehabilitation of metal-contaminated soils to extract metals and to aid the establishment of other species to enhance the phytoremediation processes.

KEYWORDS: Restoration; phytoextraction; metals; facilitation

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Testing the suitability of a carbon-storing plant for ecological restoration and rehabilitation of metal-enriched tailings

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ABSTRACT

Terrestrial life on earth depends on healthy soil to retain and release nutrients for plant growth. Plants capture carbon from the atmosphere and release it to the soil via root exudation and litter deposition. Geophytes can store carbon in living underground storage organs, which enhances soil organic carbon. In functional soils, organic carbon is essential for critical biological and physiochemical processes, and it drives many key ecosystem services. In extreme soil environments, like mine tailings, few plants survive the adverse physicochemical characteristics such as high levels of trace metals and lack of organic matter. Primary colonization of tailings mainly comprises grass species with high biomass above ground. The soil environment on tailings can be improved if we can identify species tolerant of tailings conditions and with the added advantage of storing carbon below ground. In this study we focus on an easily propagated and fast-growing geophyte, *Crinum bulbispermum* (Amaryllidaceae), that forms an exceptionally large (up to 11 kg) underground storage organ. The aim was to determine if this species can tolerate the harsh conditions on mine tailings and if the contaminated soil affects the biomass production (carbon storage ability) of *C. bulbispermum* above and below ground. A pot experiment was conducted to grow plants in tailings soils with different levels of metal concentrations. Plant development and productivity were monitored. Plants were also harvested and tested for metal uptake with ICP-MS. Results indicate that *C. bulbispermum* excludes heavy metals. However, productivity of the plants reduced significantly over one year (compared to a control) and carbon accumulation in underground storage organs were hampered. Plants became stunted and the roots were deformed, all indications that this species cannot be recommended for rehabilitation. A follow-up study will now determine whether a low microbial diversity and a lack of plant growth promoting microorganisms in these contaminated soils contribute to the poor productivity of *C. bulbispermum*.

KEYWORDS: *crinum bulbispermum*, contaminated soils, geophyte, harsh soils, soil organic carbon, underground storage organ,

Trace metal exclusion mechanisms enhance invasion of Mediterranean ultramafic habitats by *Ailanthus altissima* (Simaroubaceae)

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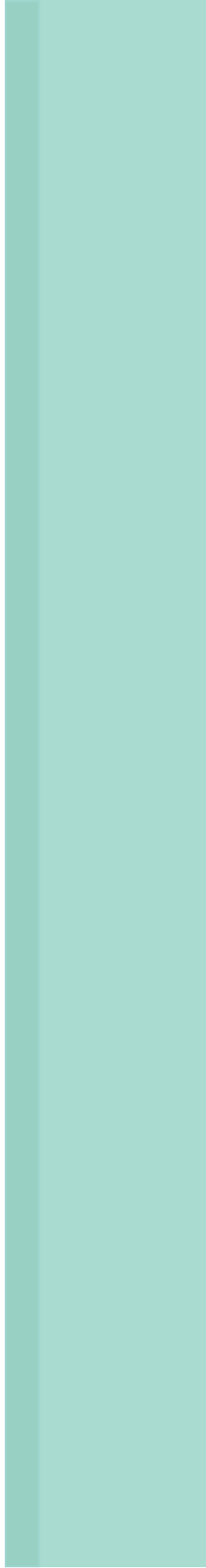
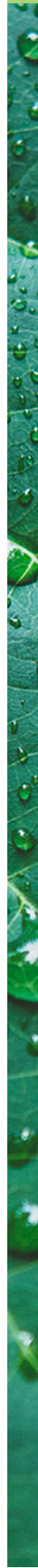
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ABSTRACT

Biological invasions are one of the major drivers of biodiversity loss worldwide. Globally, invasions by alien plants are rapidly increasing in extent and severity, leading to large-scale ecosystem degradation. *Ailanthus altissima* is one of the most invasive alien plants in Europe, being able to spread mainly in urban habitats and along transportation corridors, but also in natural habitats. Based on our field observations in Italy, this species can also invade ultramafic outcrops, which has never been reported before. This work thus aimed to evaluate the ability of *A. altissima* to invade such peculiar habitats focusing on its capacity to cope with the high concentrations of trace elements in the soil, through tolerance or accumulation/exclusion mechanisms. As comparison we used a native tree species with a similar pioneer attitude, also growing on both ultramafic and non-ultramafic soils (*Fraxinus ornus*, Oleaceae). Accordingly, we sampled five individuals of *A. altissima* and of *F. ornus* from 10 distinct populations from two different soil types: five growing on ultramafic soil and five from non-ultramafic populations as control. At each site soil and plant samples were collected to measure the concentration of trace elements (Ni, Cr, Co, Ca and Mg), and to determine the bioaccumulation and translocation factors in both species. Preliminary results indicate that concentrations of trace elements in *A. altissima* tissues are comparable to those in *F. ornus*, and always lower than in the soil. This resulted in low bioaccumulation factors and suggested trace metal exclusion mechanisms in both species. Also, Ni concentration in roots were higher than in the leaves, resulting in low translocation factors. These data suggest that *A. altissima* is unable to accumulate trace metals in its tissues but is likely to possess exclusion mechanisms that enhance its invasive ability of Mediterranean ultramafic habitats.



Session 5 - Ecophysiology, molecular biology & genetics



Addressing the diversity of the mechanisms involved in nickel hyperaccumulation

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ABSTRACT

Nickel hyperaccumulation have been described in about 500 plant species belonging to more than 50 families, mostly dicotyledons. Despite this wide diversity, most of the molecular studies on this complex trait have been focused on the hyperaccumulator species of the Brassicaceae family *Noccaea caerulescens*. Even in this model species, the demonstration that candidate genes are involved in metal hyperaccumulation is still very limited. To address the diversity in metal hyperaccumulation, we participated in a global effort to identify new hyperaccumulator species using X-ray fluorescence technology focusing on the flora of the neotropical region including Cuba [1]. The identification of nickel hyperaccumulators and closely related non-hyperaccumulator species from various plant families opens the possibility to perform comparative genomic studies to identify genes linked to nickel hyperaccumulation. Bioinformatics analysis of these results suggested that the high expression in leaves of metal transporters of the IREG/Ferroportin family is a highly conserved mechanism involved in nickel hyperaccumulation [2]. The analysis of two transporters of this family from the Cuban endemic hyperaccumulator *Leucocroton havanensis*, LhavIREG1 and LhavIREG2, revealed that the two transporters have a distinct pattern of expression and localize on the plasma membrane and the vacuole respectively. The expression of these transporters in *Arabidopsis thaliana* both increase nickel tolerance and accumulation but mobilizing distinct mechanisms. To further demonstrate the role of IREG/Ferroportin transporters and other candidate genes in nickel hyperaccumulation, we are developing *Noccaea caerulescens* as a genetic model. We recently obtained a high-quality assembly of the *N. caerulescens* genome. We now want to develop the stable transformation of this species to specifically targets candidate genes and demonstrate their precise role in nickel hyperaccumulation. Recent results will be presented at this conference.

KEYWORDS: *hyperaccumulator, rna-seq, metal transporter, ireg/ferroportin, noccaea caerulescens*

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Development of genetic tools for analysis of Ni/Zn/Cd hyperaccumulation in *Noccaea caerulescens*

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ABSTRACT

Noccaea caerulescens is a metal hyperaccumulator species in the Brassicaceae family. There are many different natural populations known, corresponding to three different ecotypes, calamine, ultramafic and non-metallicolous, analogous to the soil metal conditions they originate from. Populations have been sampled as ‘accessions’, single collections representing the original population. All accessions known so far are highly tolerant to zinc (Zn) exposure and also Zn hyperaccumulating, most accessions are also nickel (Ni) hyperaccumulating and some are cadmium (Cd) hyperaccumulating. Metal tolerance and metal hyperaccumulation traits can be genetically distinct, which means that some accessions are able to hyperaccumulate e.g. Cd, but are not tolerant, and succumb upon exposure. *Noccaea caerulescens* is a diploid species (n=7) and is fully self-compatible, unlike several other metal-hyperaccumulating *Noccaea* species, or the other well-known Zn/Cd hyperaccumulator, *Arabidopsis halleri*. This is an important advance to use this species as a model for the further understanding of the molecular mechanism of metal hyperaccumulation and -tolerance in plants. To further develop this model, we have generated a population of recombinant inbred lines (RILs), by crossing the non-metallicolous accession Lellingen, with the Zn/Cd hyperaccumulating/tolerant accession Ganges, and propagating the resulting F2 progeny to a population of 210 F7 lines. These lines have been genotyped by low-coverage whole genome sequencing, making it a suitable ‘immortal’ mapping population for genetic analysis. We will demonstrate the analysis of various plant phenotypes, related to plant growth characteristics and metal accumulation and tolerance. Furthermore, we have generated a diversity panel for *N. caerulescens*, of currently 109 accessions, representing the European germplasm of the species, which we have fully re-sequenced to generate a suitable genetic population for genome wide association studies (GWAS). We will present our first GWAS results with this population for the same traits as examined in the RIL population, and we will compare both approaches. Finally, we have set up a successful method for high efficiency transformation of *N. caerulescens*, exploiting the early flowering phenotype of an *flc* mutant we previously isolated after chemical mutagenesis [1]. Together, these three tools are expected to substantially facilitate the future research into the molecular mechanism underlying the metal hyperaccumulation phenomenon in *Noccaea* and its exploitation for phytoremediation purposes.

KEYWORDS: *Genetic analysis, genetic modification, GWAS, Zn/Ni/Cd adaptation, phytoremediation*

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Does polyploidy affect responses to Nickel in Ni-accumulating plants? Insights from the model species *Odontarrhena bertolonii* (Brassicaceae)

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ABSTRACT

Polyploidy has been recognized as a major force of plant evolution since polyploids usually display increased vigor, higher adaptive ability, and higher tolerance to extreme conditions over their diploid progenitors [1-3]. Among the various forms of stress that plants can experience, the presence of trace metals in toxic concentrations in the growth substratum is one of the most relevant ones. Among metal-tolerant plants, a few of them can accumulate the metal in the shoots to level far higher than the substrate and are termed hyperaccumulators [4]. Despite the large number of studies investigating polyploid fitness, whether the whole-genome duplication can affect growth responses, tolerance levels and accumulation ability in metal accumulators remains obscure. *Odontarrhena bertolonii* is a Ni-hyperaccumulator, restricted to serpentine soils in Tuscany and Liguria, including diploid and tetraploid populations with allopatric distribution. Three diploid ($2n = 2x = 16$) and three tetraploid ($2n = 4x = 32$) accessions were grown on the same serpentine soil and in Ni-containing hydroponic solutions and compared for growth and metal accumulation. In soil cultivation, results showed that shoot Ni concentration was similar in diploid and tetraploid plants, while the content per plant was higher in latter because of their higher biomass production. In hydroponics, tetraploid plants showed higher maximal length reached by roots and shoots and higher Ni requirement for optimal growth in respect to diploid plants, whereas Ni tolerance to both external and internal metal concentrations was similar between the two group of plants. These results suggest an effect of the genome duplication on the features of Ni-hyperaccumulation, at least in *O. bertolonii*, and recommend conducting the selection of suitable ‘metal crops’ for environmental restoration practices and field biotechnological applications at the ploidy level, when the candidate species present intraspecific polyploidy.

KEYWORDS: *diploid, tetraploid, trace metal, phytoextraction, agroming*

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Serpentine Tolerance in *Mimulus guttatus*: Gradual Acclimation to Low Ca:Mg

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ABSTRACT

In serpentine soils, the low calcium-to-magnesium ratio is detrimental to the growth and persistence of most plant species. While the exact mechanisms of tolerance are not fully understood, there appears to be ecotypic variation in *Mimulus guttatus* with adapted plants maintaining high photosynthetic rates and biomass despite elevated concentrations of Mg [1]. The mechanism providing physiological tolerance to low Ca:Mg in serpentine *M. guttatus* plant was investigated by measuring photosynthetic parameters and leaf expansion rates on excised leaf tissues, and starch and Ca²⁺ and Mg²⁺ ion concentrations following three weeks of either high (1.0) or low (0.02) Ca:Mg growth solutions. Low Ca:Mg in the disc assays reduced both photosynthesis and leaf disc expansion after one week of low Ca:Mg. However, serpentine tissues show reduced but stable photosynthetic rates after one week and a recovery in leaf tissue expansion after two weeks of exposure to low Ca:Mg conditions while values for non-serpentine tissues continued to decline. The use of DCMU, an herbicide that blocks electron transport between PSII and PSI, indicates that the light reaction is involved in the recovery of leaf tissue expansion in serpentine plants in low Ca:Mg, and the recovery is amplified by exogenous sucrose. Elevated starch concentrations at the end of the light cycle in both serpentine and non-serpentine leaves of plants grown in low Ca:Mg at one and three weeks suggests an inefficient use of carbon resources that was greater in non-serpentine plants. Both maximum photosynthetic rates and tissue expansion were negatively correlated with starch concentrations. While whole leaf and vacuole Ca concentrations did not vary between the ecotypes, serpentine leaves had higher vacuole Mg concentrations than non-serpentine leaves after three weeks of low Ca:Mg. These results demonstrate that low Ca:Mg inhibits photosynthetic rates both directly and indirectly in non-serpentine *M. guttatus* by disrupting intracellular Mg²⁺ homeostasis in non-serpentine plants. Reduced compartmentalization of excess Mg into the vacuole is likely inhibiting non-serpentine photosynthetic rates by altering the stroma pH and K transport of chloroplasts [2] and reducing leaf expansion through the increased degradation of starch and unused photosynthates that occurs with magnesium toxicity [3]. Serpentine *M. guttatus* displays an inducible tolerance to low Ca:Mg through a mechanism that maintains photosynthetic production and utilization of photosynthates necessary for growth either through gradual compartmentalization of magnesium.

KEYWORDS: *acclimation, calcium, ion homeostasis, leaf expansion, low Ca:Mg, magnesium, photosynthetic rate*

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The Role of Free L-Histidine in Ni and Zn Translocation in Hyperaccumulator and Non-accumulator Brassicaceae

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ABSTRACT

Free L-Histidine is considered to be the major Ni chelator in *Odontarrhena* Ni hyperaccumulators. During the last decades, much more information on the role of L-His, as a low-molecular-weight metal chelator, became available, not only for hyperaccumulators, but also for non-accumulator Brassicaceae. It appeared that the constitutive L-His concentrations in roots were exceptionally high not only in *Odontarrhena*, but also, seemingly genus-wide, in *Noccaea*, both in naturally Ni hyperaccumulating and naturally Zn/Cd hyperaccumulating species and accessions. Remarkably, another Zn/Cd hyperaccumulator, *Arabidopsis halleri*, showed 'normal' constitutive root L-His concentrations, but a strongly enhanced L-His concentration in its shoots, notably not in its roots, upon Zn exposure, suggesting that L-His might be essential for Zn hyperaccumulation too, at least in Brassicaceae. When exogenously supplied, via the rooting solution or foliar sprays, L-His often enhanced the translocation of Ni and, though less often, that of Zn, not only in hyperaccumulators, but also in many non-metallophytes, suggesting that the capacities for His-stimulated Ni or Zn translocation are wide-spread among Brassicaceae. Ni exposure stimulated L-His biosynthesis in a number of species, including all the *Odontarrhena* species studied thus far, and in several non-metallophytes, but never in *Noccaea*, not even in Ni hyperaccumulating species or ecotypes, nor in the Zn/Cd hyperaccumulator, *A. halleri*. Also Zn exposure enhanced L-His biosynthesis in several non-metallophytes and in *A. halleri*, though again not in *Noccaea*, not even in calamine species or ecotypes. The capacities for Ni-induced and Zn-induced L-His synthesis seem to be widely spread, both among non-metallophyte and hyperaccumulator Brassicaceae, but lacking in *Noccaea* species or ecotypes, which exhibited by far the highest constitutive root L-His concentrations among all the Brassicaceae studied thus far. Concerning the mechanism underlying the effect on the root-to-shoot translocation of Ni and Zn, it is relevant that these metals' uptake into energized root tonoplast vesicles was strongly inhibited by L-His in vesicles isolated from *N. caerulea*, but unaffected, or even stimulated, in vesicles isolated from the non-metallophyte *Thlaspi arvense*. Notably, in the latter species Ni and Zn translocation were not enhanced by exogenous L-His supply. Thus, L-His seems to enhance the translocation of Ni and Zn to the shoot through inhibiting the vacuolar retention of these metals in roots, thus enhancing their radial transport to the root stele. If so, then both Ni and Zn translocation seem to be consistently limited by these metals' vacuolar retention in roots in all the *Noccaea* hyperaccumulator species and ecotypes tested thus far, possibly also in the Zn/Cd hyperaccumulator, *Arabidopsis halleri*, and in several non-metallophytes, e.g., *A. lyrata*, *Lepidium ruderalis* and *Capsella bursa-pastoris*, but seemingly not in most of the *Odontarrhena* Ni hyperaccumulators. In *Odontarrhena*, the translocation of Ni and, possibly, Zn, was unaffected in most of the species studied thus far (data for Zn are scarce, but in *O. corsica*, there was no effect), suggesting that at least their Ni translocation capacity is generally not limited by vacuolar retention in the root, possibly because of strongly enhanced L-His synthesis under Ni exposure. It seems that translocation of Ni or Zn in *Thlaspi arvense*, as well as in many *Odontarrhena*, is not limited by their vacuolar retention in roots, but possibly, by the Ni or Zn xylem loading capacities as such. Of course, L-His might not only affect these metals' root-to-shoot transport, but also their cell-to-cell transport, and thus their distribution patterns over cells and tissues, as well as their subcellular compartmentalization in both roots and shoots, suggesting that L-His might also affect the plants' Ni or Zn tolerance capacities. However, L-His-dependent Ni tolerance has been demonstrated exclusively in transgenic *A. thaliana*, and data for Zn tolerance are overall lacking thus far.

KEYWORDS: histidine, hyperaccumulation, zinc, nickel

X-ray Fluorescence herbarium ionomics on plants from ultramafic, calcareous, and volcanic soils in Japan

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ABSTRACT

The soil type is crucially important for the concentration and solubility of essential elements required by plants, and therefore, key to the plant ionome (e.g., the totally of the mineral nutrient and trace element composition of a plant). X-ray fluorescence spectroscopy (XRF) herbarium analysis has become a powerful tool in the discovery of metal/metalloid hyperaccumulator plants. The elemental information obtained from XRF scanning of herbarium collections can also be used to study plant nutrition by creating large databases on the relationships between plants and soil. Japan is one of the world's most volcanically active countries, and approximately 30% of the surface area has soils ('Andosols') originating from volcanic ash. However, Japan also has soils with high pH, such as ultramafic and calcareous soils, but their surface area is limited, covering only 0.14% and 0.02% of Japan, respectively. Plants inhabiting in these soils are different clearly from those in other soil types and form their own vegetations. In 2019, we started XRF herbarium scanning of Japanese wild plants and collected data of over 2000 herbarium specimens as of 2022. We have used this ionome data to study the (i) sulfur and phosphorus accumulation in plants originating from Andosols and from non-volcanic soils (including ultramafic and calcareous soils), and (ii) the concentration of eight elements (potassium, calcium, sulfur, phosphorus, iron, manganese, zinc and silicon) in plants originating from ultramafic, calcareous, and other common soils of Japan (Andosols, brown forest soils and other soils). The results show that the sulfur concentrations in wild plants from Japan is higher in Andosols, but there was no difference in the phosphorus concentrations. The analysis reveals a strong correlation between sulfur and phosphorus concentrations in plants. The average phosphorus and calcium concentrations of ultramafic plants were less than half the concentrations of those of plants from other soils. The plants from other soils and calcareous soils showed a positive correlation between the concentrations of potassium-phosphorus and sulfur-phosphorus, but a statistical positive correlation between potassium-phosphorus was not found and there appeared to be a strong potassium-sulfur correlation in the ultramafic plants. The data suggests that sulfur and phosphorus have an important role in keeping a balance of cation (K^+) and anions (SO_4^{2-} and PO_4^{3-}) in plant cells and may increase the SO_4^{2-} ratio used for the counterpart to K^+ in ultramafic plants of low phosphorus status.

KEYWORDS: Andosol, ion balance, macro and micro elements, ultramafic soil, XRF herbarium ionomics

Indium Knocks at the Door: The Mechanism of Rice Inhibition by Emerging Contamination Elements

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ABSTRACT

Indium is widely used in high-tech manufacturing products and is an emerging contamination element. The presence of indium in soil inhibits shoot and root growth of rice plants, leading to yield losses. However, the underlying mechanisms remain unclear, making it challenging to design effective solutions. In this study, we investigated the comprehensive effects of excess indium on the morphological, physiological, and biochemical properties of rice (*Oryza sativa* L.). Indium treatment caused leaf necrosis and anatomical changes in the roots, particularly enlarged epidermal and exodermal layers, and thinned lignified cell wall of sclerenchyma. Indium mainly accumulated in the roots, severely restricting their growth and perturbed phosphorus, magnesium and iron homeostasis. Whole-genome transcriptomic analysis revealed that rice promptly responded to indium stress by activating genes involved in heavy metal tolerance and phosphate deficiency responses. Direct indium toxicity rather than phosphate deficiency was determined as the main factor affecting the growth of rice plants, resulting in the notable phenotypic changes we observed. Intriguingly, the application of exogenous phosphate ameliorated indium toxicity by reducing indium uptake. Our results imply that indium immobilization could be used to prevent indium toxicity in rice fields. The research on the use of phosphate fertilizers to alleviate the poisoning of the emerging polluting element indium is worthy of further investigation.

KEYWORDS: *rice, emerging contaminants, indium, heavy metal, phosphate starvation*

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The hyperaccumulator *Leucocroton havanensis* overexpress two Ferroportins/IREGs involved in nickel transportation

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ABSTRACT

Nickel hyperaccumulation is a rare and complex trait, result of the extreme adaptation of metal homeostasis networks in plants that grow in ultramafic substrates. The phylogenetic distribution of the hyperaccumulators in more than 50 families suggests this trait evolved multiple times independently from basic mechanisms involved in metal homeostasis. Constitutive high expression of IREG/Ferroportin transporters has been proposed as a convergent mechanism involved in this physiological process [1]. This study used next-generation sequencing technologies to analyze the differentially expressed genes in the nickel hyperaccumulator *Leucocroton havanensis* cultured *in vitro*. We also compared the transcriptome of *L. havanensis* and the closely related non hyperaccumulator *Lasiocroton microphyllus* from their natural environments. Arabidopsis and yeast were used as heterologous systems to study the activity and cellular localization of identified IREG transporters. Slight differences on gene expression were found between *L. havanensis* cultured with and without nickel in the media. The absence of nickel induced the expression of few genes, mainly related to biotic and abiotic stress. Using cross-species transcriptomic comparison, we identified differential expressed contigs coding for members of the IREG/FPN family. *L. havanensis* highly expressed two IREGs transporters compared to *L. microphyllus*. Transgenic plants overexpressing *LhIREG1* and *LhIREG2* showed an increased tolerance to elevated concentrations of nickel. The two IREGs are localized in different membranes. Heterologous expression in yeast demonstrated a nickel transport function for both transmembrane proteins. These results support the hypothesis that IREG/Ferroportin transporters emerged as a mechanism involved in nickel hyperaccumulation.

KEYWORDS: *Hyperaccumulation; rna-seq; metal transporter; ireg/ferroportin*

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Leaf elemental composition of species growing on contrasting soils in two adjacent rainforest: serpentinized ultramafic versus volcano-sedimentary rock

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ABSTRACT

The flora of New Caledonia is renowned as one of the world's most significant biodiversity hotspots. The contrasting soil conditions characterizing this small archipelago are known to largely influence species local diversity and distribution. Because the difference between soil chemistry is likely to cause variation in the leaf elemental composition, we wanted to test how different soil condition affect plant community and leaf nutrient concentration. We focused on two adjacent forests, of similar physiognomy, growing on serpentinized ultramafic rock and on siliceous volcano-sedimentary rock. Both soil strongly differed in their pH, metal element concentration (Al, Ni, Mn), and ions exchange capacity. The two adjacent forests have a rich and diverse flora, and share a relatively high fraction of species (35-42 %). The total vascular flora had a highest similarity compared to the tree flora only. The foliar element concentrations of 30 tree species (4 to -9 samples per species) that grow on both soil types were analysed. The differences in leaf mineral composition were more important between species than for similar species growing on different soils. Globally, on both soils, the leaf element profile reflects low availability in major elements: nitrogen, phosphorus, calcium, and potassium. Despite high total or DTPA-extracted manganese contents in ultramafic soils, the foliar manganese concentrations are significantly higher over volcano-sedimentary rocks. Foliar nickel levels are, as expected, higher on ultramafic substrates and aluminum concentration is higher on volcano-sedimentary rocks. Finally, the major difference in leaf elemental concentration concern metal elements (Ni, Mn, Al) while major nutrients vary in much lower proportion between the two soil types, suggesting a thight regulation of major nutrient compared to trace element.

KEYWORDS: *Leaf mineral composition, metal concentration, New Caledonia, rain forest, species composition, soil chemistry, ultramafic soils.*

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

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ABSTRACT

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

KEYWORDS: Nickel, *Bornmuellera emarginata*, Physiology, Nickel Agromining

Assessing Nickel Tolerance and Accumulation in the Central American Hyperaccumulator *Blepharidium guatemalense* Using Hydroponic Systems

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ABSTRACT

Blepharidium guatemalense Standl. is a Central American tree in the Rubiaceae (coffee family), recently discovered to hyperaccumulate nickel (Ni). Exceptionally high foliar Ni concentrations, exceeding 4% of dry mass, have been measured under field conditions in Mexico and Guatemala [1] and after experimental cultivation in Ni-amended soils [2]. The purposes of the present study were to investigate whether *B. guatemalense* can be grown in hydroponic culture solution in an environmental chamber, and to measure its responses to a range of Ni concentrations in the culture medium. Seeds of *B. guatemalense* were germinated on wet sand, and 20 seedlings were transferred to hydroponic boxes with their roots immersed in 10%-strength Hoagland nutrient solution, amended to produce Ni concentrations ranging from 0 μM to 300 μM . After 5 months of growth, we measured the dry weight of the roots and shoots and performed atomic absorption spectroscopy on digested tissue samples to determine the concentration of Ni in each plant's roots and leaves. There were no significant differences between treatment groups in the masses of shoots and roots, indicating no symptoms of Ni deficiency at low concentrations and no symptoms of Ni toxicity at high concentrations. Shoot and root Ni concentrations increased with solution Ni concentration, exceeding the 1,000 mg kg^{-1} D.W. foliar concentration threshold for hyperaccumulation even in solutions with low Ni concentrations, and reaching a maximum foliar Ni concentration over 16,000 mg kg^{-1} (1.6 %) in the 300 μM solution. A few plants were not harvested but cultured for an additional 2 months in elevated Ni concentrations up to 10 mM; they continued to show vigorous growth. Sample sizes in our study were small due to Covid-related travel restrictions. However, we have shown that this tropical tree can be grown successfully in hydroponic solutions, providing an additional tool for the study of its physiological responses. *Blepharidium guatemalense* has been suggested as a potential candidate for agromining of Ni in tropical areas [2], making it an attractive subject for further investigations.

KEYWORDS: *hyperaccumulator, nickel, metal tolerance, agromining, hydroponics, serpentine*

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Exclusion and Accumulation: Metallophytes Employ Both Mechanisms to Survive on Serpentine and Mining Habitats of Sekhukhuneland, South Africa

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ABSTRACT

Worldwide, serpentine outcrops are often disturbed by mining activities that not only destroy the habitats of highly specialized flora but also cause widescale pollution of the environment with potentially toxic metals (PTMs). Under such circumstances, it is important to evaluate if anthropogenic factors (i.e., land use practices) have a greater influence on metal distribution in the soil-plant system compared to natural factors (i.e., local topography and geology). Localities in Sekhukhuneland, an ultramafic region of South Africa, were selected along an altitudinal gradient (topography: upper slope, footslope, valley and valley bottom) and a land use profile (rangelands, gardens, tailings and wastelands) to investigate the distribution of Co, Cr, Cu, Fe, Mg, Mn, Mo, Ni, Sr and Zn in surface soil and plant leaf tissue. Plant life form was considered as an additional factor to evaluate PTM accumulation in leaves. Findings revealed higher concentrations of Cr and Ni in soils throughout the catena compared to WHO standards. Other than Mg, abundant soil metals (Cr, Fe, Mn, Ni) were excluded (bioaccumulation factor, BAF < 1) by all plant species. Conversely, Co, Cu, Mg, Mo, Sr and Zn were accumulated (BAF > 1) by a high percentage of evaluated plants (74%), mostly indigenous (83%). Grasses, taller forbs, dwarf shrubs and shrubs, in that order, showed the greatest accumulation abilities. Distribution of PTMs in soils and plant leaves differed among topographic positions, but was non-significant. Across land uses it differed significantly, indicating anthropogenic interference as a predominant determinant of PTM enrichment of soil and subsequent metal uptake by plants. Plants differed in their response strategy to metalliferous soils of comparatively less-disturbed and extremely polluted mining sites, but primarily excluded abundant soil metals. These traits make these species valuable for phytoremediation and rehabilitation of metal contaminated sites, as they are unaffected by metal-rich substrates.

KEYWORDS: *Bioaccumulation, excluders, metallophytes, phytoremediation, tailings*

Ion profiles of plant species distributed in two contrasting soils: serpentine soil and volcanic acid soil, western Japan

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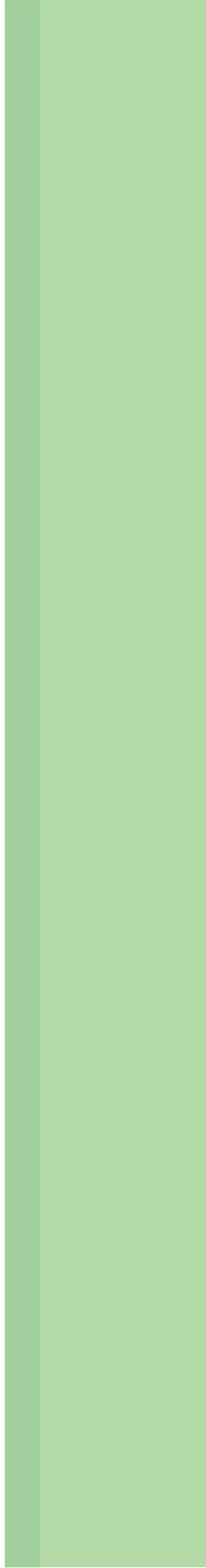
ABSTRACT

Plants have adaptations that help them to survive and grow in certain environments. Regarding pH, there are two contrasting soils in Japan: serpentine soils and volcanic acid soils (hereinafter referred to as solfatara fields). Serpentine soils are characterized by a pH of 6–8, high Mg and Ni, and low Ca, N, P, and K contents. The severely deficient levels of N, P, K, and Ca lead to serpentine soils being inhospitable to most plants. Solfatara fields, areas surrounding fumaroles near hot springs or volcanoes, are also characterized by extreme environmental conditions such as low soil pH and high Al contents. Some specific plant species (serpentine and solfatara plants) found in serpentine soils and solfatara fields, respectively, may develop strategies to cope with stresses in these areas in Japan. This study aimed to investigate the ion profiles of specific plants found in serpentine soils and solfatara fields and to discuss the mineral accumulation properties in the plants. We harvested mature leaves from specialists (*Saussurea modesta* and *Berberis amurensis*) and a generalist (*Thymus quinquecostatus*) plant from serpentine soils in Hiroshima, and a specialist (*Fimbristylis dichotoma* subsp. *podocarpa*) and generalist (*Lycopodium cernuum*) from solfatara fields in Kyushu, and generalists (*Miscanthus sinensis*, *Smilax china* and *Clethra barbinervis*) growing in both areas. We also collected generalists in normal soils in Hiroshima (not serpentine soils or solfatara fields). Rhizosphere soils of *M. sinensis* were collected from both serpentine soils and solfatara fields. Dried plant samples were digested with HNO₃-H₂O₂ and used for mineral analysis using inductively coupled plasma mass spectrometry (ICP-MS). Soil pH (H₂O) was measured using a pH meter. The average soil pH of *M. sinensis* was 6.02 and 3.36 in serpentine soils and solfatara fields, respectively. Mg concentrations tended to be higher in serpentine than in solfatara plants. In contrast, concentrations of Al, Ca, and Fe tended to be higher in solfatara than serpentine plants. However, the value was lower than that of accumulator plants. The Al concentrations between plants from solfatara and normal plots were similar. *L. cernuum* had high levels of Al, similar to that of known Al-accumulators. The Al concentrations of other plants were low, suggesting that they had a strategy to avoid Al accumulation in their tissues. We confirmed that the soil influenced the concentration of elements in the plant. However, we also found that the characteristics of element absorption differed among plant species. Furthermore, it can be suggested that plants growing in both serpentine soils and solfatara fields may have adapted to a wide range of environments.

KEYWORDS: Serpentine plants, solfatara fields, ultrabasic rock, low pH, Al-accumulators



Session 6 - Metal hyperaccumulation discovery & monitoring



The Discovery and Global Distribution of Hyperaccumulator Plants

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ABSTRACT

As analytical chemistry developed throughout the 20th Century, information was acquired on the normal concentrations of many trace elements in plants, including Zn, Cu, Co, Ni, Cd, Pb, As and Se. Abnormally high concentrations of one of these elements were occasionally reported. In some cases, these were in plants growing on highly metalliferous substrates near mining or smelting operations; others were from undisturbed natural metal-rich soils, such as those of ultramafic regions and outcrops rich in chalcophile elements. Although an instance of extreme Zn accumulation was found in the 19th Century, observations of high concentrations of the other elements above came to light at various times between the 1920s and 1970s.

Extreme Ni accumulation in plants from ultramafic soils was recorded sporadically during the period 1948-1975. This led to more concerted analytical work, using fragments of herbarium specimens as well as fresh material collected in the field. A focus on Ni accumulation was driven partly by the fact that nickel-rich ultramafic soils are globally widespread. In some regions they extend over several hundreds of km², and can be notable for hosting numerous endemic species. The term hyperaccumulator was introduced in 1976 to signify a species exhibiting a concentration that can be hundreds or even thousands of times greater than that usually found in plants on most common soils. Attempts have been made to specify concentration criteria for various elements in plant tissue that enable a species to be regarded as a hyperaccumulator. Such criteria are to some degree arbitrary, but are intended to focus attention on species in which some unusual features of plant metabolism must be present.

The interest in hyperaccumulators has prompted many new investigations on different aspects of the biota of a variety of metalliferous soils in many parts of the world. The search for new examples of hyperaccumulation continues, aided in part by non-destructive X-ray fluorescence scanning of herbarium specimens. Ultimately this needs to be supported by rediscovery of species of interest in the field, and by detailed plant and soil sampling to better understand the factors governing the level of metal uptake. More than 700 species that qualify as hyperaccumulators under strictly defined criteria are now regarded as a resource for many types of fundamental scientific investigation (plant systematics, ecophysiology, biochemistry, genetics and molecular biology) and for applications such as phytoremediation and agromining. Because many of these species are relatively rare and are of restricted distribution, conservation issues are often important.

KEYWORDS: metalliferous soils; nickel; hyperaccumulation

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Exploring the associations between soil microbiome, root architecture, and Zn & Cd accumulation in *Arabidopsis halleri*: results from a transplant experiment in rhizoboxes

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ABSTRACT

Metal-hyperaccumulating plants possess complex traits that allow them to thrive in soils with high concentrations of trace metal elements (TME). Their TME hypertolerance and hyperaccumulation capacities have been intensely studied from physiological, evolutionary, and ecological perspectives [1]. Little is known, however, about root system development in TME enriched vs natural soils, and about the associated rhizosphere microbial communities that may facilitate TME uptake and tolerance [2]. The goal of this study was to characterize the diversity of the soil microbial communities, plant performance, and metal uptake by non-metallicolous (NM) and metallicolous (M) populations of *Arabidopsis halleri* in metalliferous and non-metalliferous soils, thereby following a reciprocal transplant experiment design. We quantified temporal changes in root system architecture using a combination of root phenotyping in rhizoboxes and multi-temporal digital imaging. Additionally, we monitored shoot and root growth, characterized the diversity and structure of *A. halleri* rhizosphere-influenced soil microbial communities, and assessed plant-microbial interactions using combinations of various plant traits and soil biogeochemical characteristics. Our results indicate that differences in *A. halleri* TME-hyperaccumulation capacities are primarily driven by the plant population. Temporal observations of plant development allowed to detect dynamic variability in growing strategies between populations and were influenced by both edaphic type (NM vs. M) and seed sterilization. Further, shifts across populations and soil treatments were observed for the soil microbial network analyses, resulting in 71 unique keystone bacterial/archaeal (57) and fungal (14) phylotypes. These critical microbes had strong associations with Zn and Cd bioaccumulation factors - suggesting that key species interactions among diverse groups of soil microbes further shape plant TME tolerance and uptake.

KEYWORDS: *hyperaccumulation; hypertolerance; phytoremediation; plant-microbial interactions; rhizobox; root architecture; transplant experiment*

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Metal Hyperaccumulators of Mexico and Central America: A Survey of Recent Progress

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ABSTRACT

Until 2016, no hyperaccumulators of heavy metals were known from Mexico or Central America. At the 2017 ICSE in Albania, McCarthy et al. reported hyperaccumulation in four species of *Psychotria* (Rubiaceae), ranging from Mexico to Ecuador, based on XRF scanning of herbarium specimens [1]. Two were previously known from Cuba, but two were newly identified hyperaccumulators. Interestingly, the Mexican hyperaccumulators were from areas in the states of Veracruz, Tabasco, and Chiapas that are not geologically mapped as ultramafic. Field excursions in 2017 and 2018 visited the sites. In Veracruz, weathering of basaltic bedrock produces soils with moderate Ni concentrations. On these soils, *Psychotria papantlensis* hyperaccumulates Ni, but more remarkably, some individuals simultaneously hyperaccumulate Ni and Zn, often with high Al/Cd/Co concentrations as well. Fieldwork revealed another new Ni hyperaccumulator, *Orthion veracruzense* (Violaceae). Studies in Chiapas and Tabasco suggested that Ni enrichment of the soils may result from transport of ultramafic minerals by water or volcanic ash. The hyperaccumulator status of *Psychotria costivenia* and *P. lorenciana* was confirmed in the field, and other new hyperaccumulators were found, including *Blepharidium guatemalense* (Rubiaceae) [2] and a second species of *Orthion* – *O. subsessile*. Subsequent herbarium scanning at MO, P, MEXU, USCG, and UVAL revealed that six species of Neotropical *Orthion*, plus the closely related *Mayanaea caudatum*, all include specimens with foliar Ni exceeding 1000 µg g⁻¹. The most extensive ultramafic deposits in Central America occur in the Motagua fault zone of Guatemala. Examination of herbarium specimens revealed two additional hyperaccumulators in this region: *Arachnothryx linguiformis* (Rubiaceae) and *Chionanthus panamensis* (Oleaceae) [3]. Field and herbarium work in Guatemala in 2019 allowed further study of these species, which occur in a diverse community of hyperaccumulators along with *B. guatemalense*, *O. subsessile*, and *P. costivenia*. At least 14 species of hyperaccumulators are now documented in Mexico and Central America.

KEYWORDS: *hyperaccumulate; metallophyte; nickel; ultramafic; serpentine; ophiolite; Mexico; Guatemala*

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Trace element hyperaccumulator plant traits: a call for trait data collection

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ABSTRACT

Hyperaccumulator plants exhibit extreme ecophysiological characteristics, which make them valuable for conservation and suited for phytoremediation¹. Understanding their ecological strategies might help identify the species and functions to be fostered in phytoremediation, restoration, and conservation projects for metalliferous sites². Here, we identified the hyperaccumulator species in the worldwide plant trait database TRY and cross-referenced these trait syndromes associated with the ability of plants to concentrate metals. We collected three traits (leaf area (LA), specific leaf area (SLA) and leaf dry matter content (LDMC)) on all available species on TRY (n= 4523), from which 20 were identified on the global hyperaccumulator database. This allows us to link trace element hyperaccumulation with broader plant ecological strategies, such as competitive, stress-tolerant and ruderal strategies (CSR) or resources acquisition ones. With these primary data, hyperaccumulators seem to have smaller leaves and poor competitive ability compared to non-hyperaccumulator species. Contrary to expectations, we found no indication of hyperaccumulators being more resource-conservative on the leaf economics spectrum. However, these data remain fragmentary as only 2.7% of hyperaccumulators have their traits published in the TRY database. We thus proceed to a systematic review to retrieve around 40 additional new hyperaccumulator species present on TRY and call for an international collaborative sampling effort to measure traits in more hyperaccumulator species. The recent development of trait-based models to construct plant communities providing optimal ecosystem services (e.g., phytoremediation, restoration)³ requires further research to identify predictable trait-service relationships. This research provides a first synthesis of the research advancement on hyperaccumulator traits, as well as a first synthesis analysis of their strategies at a global scale.

KEYWORDS: *csr strategies, functional traits, hyperaccumulation, metallophytes, plant functions, synthesis analysis*

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XRF ionomics of herbarium specimens from the Balkan Peninsula

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ABSTRACT

Herbaria are important source of information for various taxonomic and biogeographic studies. Novel methodologies are enabling new ways for the non-destructive analysis of herbarium material, including the analysis of plant ionomes using portable X-ray fluorescence (XRF) instrumentation. The analysis of the ionome i.e., the elemental composition of a plant, can contribute to the understanding of the homeostasis of elements in plant tissues, as well as led to the identification of the hyperaccumulation of certain elements, which is a very rare phenomenon in the plant world. The diverse geology of the Balkan Peninsula with a significant occurrence of nickel-bearing ultramafics contributes to the occurrence of a large number of hyperaccumulators of this element. In addition to nickel, hyperaccumulation of zinc, arsenic and thallium has also been detected in taxa from this area. The most important collections of plant specimens from this region are deposited in the Herbarium of the University of Belgrade (BEOU), and in the collection of the Natural History Museum in Belgrade (BEO). In order to analyze the ionome of plants, and potentially discover new hyperaccumulators, the collections of these two herbaria were systematically scanned with XRF, paying particular attention to the representatives of the Brassicaceae, Violaceae, Plantaginaceae and Euphorbiaceae families. Of the 3000+ herbarium specimens scanned, hyperaccumulation of Ni was confirmed in a number of taxa of the Brassicaceae family (*Noccaea*, *Bornmuellera*, *Odontarrhena*), as well as in the genus *Viola* (*Viola dukadjinica*; Ni > 1,700 mg kg⁻¹). Hyperaccumulation of Zn was detected in species of the genera *Noccaea* and *Cardamine*, whilst unusual accumulation of manganese was recorded in species of *Euphorbia* (*E. glabriflora*; Mn > 4,500 mg kg⁻¹ in several specimens). According to the results of the XRF scanning, *Cardamine waldsteinii* is a potential new Zn hyperaccumulator (Zn > 3,000 mg kg⁻¹), but only strong accumulation was confirmed by the analysis of living material. Among the most interesting results of this study is the simultaneous hyperaccumulation of Ni and Zn, that has been detected for the first time in several specimens of *Noccaea praecox*. Hyperaccumulation of thallium and extremely high arsenic concentrations were detected in representatives of *Viola*, endemic to the Allchar site located in North Macedonia. The Tl concentrations in several specimens of *Viola arsenica* exceeded 16,000 mg kg⁻¹. Further systematic scanning of herbarium collections in the Balkan Peninsula could contribute to the discovery of new hyperaccumulator plants with the potential for application in biotechnology, as well as to gaining a better understanding of the complex phylogenetic relationships of certain genera with significant diversity in the Balkans.

KEYWORDS: XRF, herbaria, hyperaccumulation, ionomics

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Nickel accumulation in plants from Shebenik mountain massif

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ABSTRACT

Hyperaccumulators are unusual plants that accumulate particular metals or metalloids in their living tissues to levels that may be hundreds or thousands of times greater than is normal for most plants. Hyperaccumulative plants are of practical importance as they are used in soil rehabilitation technologies. Hyperaccumulators are exceptional models for fundamental science to understand metal regulation including the physiology of metal uptake, transport and sequestration, as well as evolution and adaptation in extreme environments. The aim of this study was to determine; (i) the different taxa of nickel hyperaccumulator plants present in Shebenik mountain massif; evaluation of Ni concentration in different collected taxa in different elevation; (ii) relationships between metal in the soil and Ni accumulation by different taxa or populations. Morphological and karyological analyses of material from field collections across serpentine/ultramafic outcrops of the Shebenik Mountain allowed us to determine those taxa: *Odontarrhena chalcidica*, *Odontarrhena smolikana* subsp. *Glabra*, *Odontarrhena rigida* and *Thlaspi* ssp. Analysis of element concentrations in aerial parts of plant samples, soil samples, DTPA-extractable Ni and determination of biomass production allowed evaluation of taxa efficacy in hyperaccumulating Ni and using in phytoextraxtion/phytomining technologies.

KEYWORDS: *Hyperaccumulators, Nickel, Shebenik mountain, serpentine, agromining*

Biogeochemistry of the Serpentine of Rhodes and Cyprus

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ABSTRACT

Ultramafic biogeochemistry of the islands of Rhodes and Cyprus has not previously been discussed at ICSE conferences. In this presentation we note the incidence of ultramafic rocks and soils on both islands, the occurrence of endemic plant species, typical soil and plant elemental compositions and the presence of nickel hyperaccumulating species. Areas of serpentine soils on Rhodes are few, small and not strongly marked in relation to their surroundings. The very rare serpentine endemic *Alyssum pogonocarpum*, found on Rhodes and in SW Turkey, is of some interest. By comparison, the ultramafics of Cyprus have been well studied, and the Troodos massif in particular is of global geological significance. Ni hyperaccumulation in five species from Cyprus (four in *Alyssum* – now transferred to *Odontarrhena*, and one in *Thlaspi* – now transferred to *Noccaea*) was found by analysis of a few herbarium specimens in 1979-83, but little further work has been done on these. We discuss the distribution of *Noccaea cypria*, *Odontarrhena akamasica*, *O. chondrogyna*, *O. troodi* and *O. cyprica*. The last of these occurs also in SW Turkey, but the other species are endemic to Cyprus. Further elemental data is now available on all these species and their associated soils, as well as for some other plant species from the same areas. Ni hyperaccumulation by the above species is confirmed, but *A. pogonocarpum*, from sect. *Alyssum* of the genus *Alyssum*, is not a hyperaccumulator. Conservation issues are noted, and aspects of generic nomenclature (*Alyssum* vs. *Odontarrhena*) are discussed.

KEYWORDS: Rhodes, Cyprus, *Alyssum*, *Odontarrhena*, nickel hyperaccumulators

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Element Accumulation by the Holoparasitic Species *Cuscuta planiflora* Ten. from Serpentine in Bulgaria

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ABSTRACT

The holoparasitic species *Cuscuta planiflora* Ten. (Convolvulaceae) parasitizes on different flowering plants. It is a species found to parasitize on the Ni hyperaccumulator *Odontarrhena muralis* (Waldst. & Kit.) Endl. in Bulgarian serpentine areas in the Eastern Rhodope Mts. This study aims to investigate host/parasite relationship to evaluate: (1) the metal accumulation in different parts of the parasite and the host plant, and (2) a potential threat to Ni phytomining/agromining by suppressing the growth of the plant hyperaccumulator. Elemental concentrations in soil and plant (stems, leaves, inflorescences) samples were measured for the organs in two hosts: *O. muralis* and the widely spread on serpentine non-hyperaccumulator *Sanguisorba minor* Scop., as well as for the stems and inflorescences in *C. planiflora*. The elemental concentrations for Ca, Mg, Fe, Ni, Cr, Co, Zn, Cu, Mn, Na, Al were measured after microwave digestion using ICP-OES under optimal instrumental parameters. Individuals of *O. muralis* and *S. minor*, both infected and non-infected, showed slight differences in their composition of the essential elements (P, K) and of some micronutrients. The infected individuals of *O. muralis* showed lower Ni concentrations in stems, leaves and inflorescences relative to the non-infected ones. *Sanguisorba minor* is an excluder plant growing on serpentine soils with low Ni concentrations in its tissues. The parasite has higher P, K and lower Ca, Mg, Ni, Zn, Co, Mn and Al concentrations than the host. The highest Ni concentration measured in the stems of *C. planiflora* was 70 mg kg⁻¹ parasitizing on *O. muralis*, whereas that from *S. minor* was 30 mg kg⁻¹. The transfer of the chemical elements from the host to the parasite is element specific. The hyperaccumulation of Ni by *O. muralis* did not prevent attack by *C. planiflora*. After infection, there was a decline in Ni concentration and the dry weight of all organs of the host. The infection of *O. muralis* is the reason for the measured reduced biomass yield. This infection could be a potential threat to the use of *O. muralis* for Ni agromining.

KEYWORDS: *Cuscuta planifolia*, holoparasite, serpentine, metal accumulation, Ni hyperaccumulator, biomass

Assessing the metal accumulation capacity of plants to optimize phytoextraction of mine tailings in arid and semi-arid ecosystems

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ABSTRACT

Phytoextraction is a promising strategy for remediation of metal-contaminated mine tailings [1]. This technology capitalizes on the ability of plants to effectively harvest metals from soil through root uptake and transport into above-ground tissues. However, there is a lack of information on plant metal accumulation in arid and semi-arid climatic regions, which presents a challenge for the selection of plant species for phytoextraction [2]. To address this gap, we are conducting extensive in-situ surveys of legacy mine sites and natural outcrops in Arizona, using a portable X-ray fluorescence analyzer (pXRF). This tool rapidly characterizes metal accumulation in plant tissues, spanning across various plant families. To date, using this technology, we have been able to identify numerous species with a significant capacity for uptake of Cu, Se, Zn and Re. Further, plant-derived metal complexes can be subsequently used as “ecocatalysts” to replace traditional catalyst in organic chemistry reactions and industrial production of materials (e.g., bioinspired cosmetics, fragrance, or pharmaceutical industry). Consequently, innovative methods are being developed to expand the scope of plant identification research, encompassing the use of Micro X-ray fluorescence spectroscopy to analyze metal allocation in plant tissues *ex-situ*. This research not only broadens our understanding of strategies underlying plant metal adaptation in arid regions but also supports the mining industry in sustainability by providing technology for metal recovery from mine tailings.

KEYWORDS: *phytoextraction, phytomining, phytoremediation, hyperaccumulation, xrf analysis, ecocatalysis, heavy metals, mine tailings*

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Atomic Spectroscopic Analysis of Macronutrients and Trace Elements in Potential Hyperaccumulators from Sabah

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ABSTRACT

Hyperaccumulators species are widely distributed on ultramafic lands where it is capable to accumulate extremely high concentration of heavy metals such as Nickel (Ni), Cobalt (Co), Manganese (Mn) and etc. The accumulation of metals in the plants often occur in the roots, stems and leaves without showing any signs of toxicity. Currently, the study of hyperaccumulating plants as new resources in the development of novel plant-based technologies for the treatment of polluted sites and extraction of rare metal is extensively carried out in Sabah where the study has been proven to be successful in USA, Europe and China since 1980s (van der Ent *et al.*, 2018). However, the accurate number of identified hyperaccumulators in Sabah and related information such as elemental distribution is not known due to lack of fundamental studies regarding hyperaccumulators in Sabah, Malaysia. The aim of the study is to suggest hypernickelophores as tropical metal crop candidate for agromining through evaluation of macronutrients and trace elements in identified hyperaccumulators by using atomic spectroscopic analysis. 8 species of identified hyperaccumulating plants (*Psychotria sarmentosa*, *Glochidion* sp. 'bambangan', *Rinorea, bengalensis*, *Rinorea javanica*, *Actephila alenbakeri*, *Walsura pinnata*, *Xylosma luzonensis* and *Mischocarpus sundaicus*) were collected from serpentinite area in Kinabalu Park Reserved Forests which are Pig Hill Mesilau, Garas Hill Ranau, Serinsim Sub-station Kota Marudu and Monggis Sub-station Ranau. The study is carried out by determining the concentration of macronutrients and trace elements such as sodium (Na), calcium (Ca), potassium (K), nickel (Ni), cobalt (Co) and Manganese (Mn) of identified hyperaccumulators and the soils where they grow by using Atomic Absorption Spectrophotometer (AAS) prior to samples preparation and wet digestion of the samples. 4 hypernickelophores are found in this study which are *G.* sp. 'bambangan', *R. bengalensis*, *R. javanica* and *P. sarmentosa* with the accumulation of Ni as much as 10 784, 13 196, 13 780 and 17 085 $\mu\text{g g}^{-1}$, respectively. However, the accumulation of Ni in *R. javanica* from previous study is only 9680 $\mu\text{g g}^{-1}$ which is below the threshold limit for hypernickelophore, 10 000 $\mu\text{g g}^{-1}$ (van der Ent *et al.*, 2017). The accumulation of other trace elements (Co and Mn) is unremarkable as the amount is low ranging from 5 to 443 $\mu\text{g g}^{-1}$. Apart from that, the accumulation of Ca is the highest in the leaf of collected samples compared to ranging from 5625 to 20 7997 $\mu\text{g g}^{-1}$ followed by K in the concentration of 993 to 18 990 $\mu\text{g g}^{-1}$. High concentration of Ca and K in the samples is due to the relation of these elements in various physiological mechanisms such as water conservation strategy (van der Ent, 2013). The accumulation of Na in samples is low ranging from 50 to 235 $\mu\text{g g}^{-1}$. Analysis of soil elemental concentrations shows that the concentration of Na, Ca, K, Ni, Co and Mn is ranging from 114 to 470 $\mu\text{g g}^{-1}$, 2495 to 5606 $\mu\text{g g}^{-1}$, 51 to 649 $\mu\text{g g}^{-1}$, 432 to 4265 $\mu\text{g g}^{-1}$, 130 to 937 $\mu\text{g g}^{-1}$ and 156 to 1335 $\mu\text{g g}^{-1}$ while the pH of soil samples is common for serpentine soil which is ranging from 6.56 to 7.48. Therefore, further study should be carried out for *G.* sp. 'bambangan', *R. bengalensis*, *R. javanica* and *P. sarmentosa* in order to determine the most suitable agronomic practices so the hypernickelophore can be utilized in the application of phytoremediation and phytoextracting which is still new in Malaysia, particularly Sabah.

hypernickelophore; serpentinite; phytoremediation; phytoextraction

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Does metal hyperaccumulation influence leaf traits? Morphological and anatomical investigation of the ultramafic flora of New Caledonia

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ABSTRACT

Throughout the world, ultramafic soils support distinct plant communities, most often composed of endemic species. This is because the physical conditions and chemical composition of ultramafic soils impose drastic conditions for plant growth, and specific mechanism of species tolerance and adaptation to ultramafic soils. These edaphic conditions, referred to as the "serpentine syndrome", are low moisture and nutrient availability, cation imbalance, and often high trace metal concentrations. Thus, it is reasonable to expect that plant communities in ultramafic soils share a set of functional traits, specifically traits that confer stress tolerance, low resource acquisition, and metal avoidance. Specific studies of plant functional traits in relation to ultramafic soils are scarce, but tend to show that species share reduced leaf size, low specific leaf area, slow growth rate, and sclerophyllous leaf anatomy, among other common characteristics. Among ultramafic plant communities, hyperaccumulator plants represent an extreme physiological strategy to cope with high levels of trace metals, as these species store high concentrations of trace metals (e.g., Ni, Mn) in their aerial tissues and especially in their leaves. It has been suggested that hyperaccumulation serve different functions, including osmotic regulation under water stress, increasing plant defense or enhancing plant growth. Here, we aim to analyze the functional impact of hyperaccumulation and assess whether trace metal hyperaccumulation is reflected in functional leaf traits. We compare a set of morphological (SLA, LDMC, leaf thickness and size) and anatomical (epidermis and cuticle thickness, proportion of different tissues) traits of hyperaccumulating (for Mn and Ni) and non-hyperaccumulating species. The species were selected from the same genera and growing in sympatry in the same environment, on an ultramafic soil in New Caledonia. We found that all species share sclerophyllous traits and occupy a conservative resource economy in the leaf economic spectrum, reflecting their resource conservation strategy to establish on ultramafic soils. With the exception of leaf cuticle thickness, we found no significant impact of hyperaccumulation on leaf traits.

KEYWORDS: *hyperaccumulation, functional trait, leaf anatomy, metal concentration, New Caledonia, serpentine soil, ultramafic soils.*

Herbarium and Field Studies of Nickel-Hyperaccumulating Plants from Ultramafic Soils in Guatemala

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ABSTRACT

[Recent discoveries of metal-hyperaccumulator plants in Central America [1, 2] prompted further exploration of ultramafic areas in this region. The most extensive serpentine and nickel-laterite deposits in Central America occur in eastern Guatemala, along the Motagua fault zone at the boundary of the Caribbean and North American plates [3]. We used X-ray fluorescence analysis to measure elemental concentrations in herbarium specimens at the Missouri Botanical Garden that had been collected in this zone and belonged to genera known to include hyperaccumulators, according to the global hyperaccumulator database [4]. Using this search strategy, we discovered two species not previously known as hyperaccumulators. *Chionanthus panamensis* (Oleaceae) is a small tree ranging from Panama to southern Mexico. It is a facultative hyperaccumulator, with low Ni concentrations across most of its range, but foliar Ni concentrations up to 5000 $\mu\text{g g}^{-1}$ DW on metalliferous soils in Guatemala and Mexico. *Arachnothryx linguiformis* (Rubiaceae) is a shrub with foliar Ni exceeding 20,000 $\mu\text{g g}^{-1}$ in some specimens. It is an obligate hyperaccumulator, endemic to ultramafic outcrops around Lake Izabal, Guatemala, and is the first species in its genus (recently segregated from *Rondeletia*) known to hyperaccumulate any element. During subsequent fieldwork in Guatemala, we could locate only one living example of *C. panamensis*, with foliar Ni concentration of 6,800 $\mu\text{g g}^{-1}$. In a sample of 14 field-collected *A. linguiformis* plants, foliar Ni concentrations ranged from 1500 to over 13,000 $\mu\text{g g}^{-1}$. Soil samples were collected under each plant; total Ni ranged from 900-10,700 $\mu\text{g g}^{-1}$, whereas DTPA-extractable Ni ranged from 300-4200 $\mu\text{g g}^{-1}$. Despite this variability in plant and soil Ni concentrations, there was no statistically significant correlation between foliar Ni and either measure of soil Ni. Discovery of new hyperaccumulators may be important in the development of biotechnology for phytoremediation and phytomining, and represents a useful research model in plant ecology, physiology, and conservation.

KEYWORDS: *hyperaccumulator; nickel; serpentine; ophiolite; Guatemala; Arachnothryx; Chionanthus*

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Hyperaccumulation of Ni and Zn in *Noccaea* herbarium specimens

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ABSTRACT

There is a growing interest in the discoveries of new hyperaccumulating species, and at the same time new methods for their discovery are being developed. X-ray Fluorescence Spectroscopy has been shown to be suitable for non-destructive elemental profiling of plant specimens. Its use in herbarium collections is of particular importance. Herbaria are a valuable source of information not only on species distribution but also on other plant traits, such as the potential for hyperaccumulation. Besides *Odontarrhena*, the genus *Noccaea* includes the largest number of hyperaccumulator species within the family Brassicaceae. It is also one of the genera with the highest number of known hyperaccumulators of Ni. Elemental profiles of herbarium specimens of *Noccaea* species were analyzed using a portable XRF instrument in the vascular plant collection of the National Museum of Natural History in Paris (MNHN). A total of 632 specimens, representing 53 taxa within this genus, were scanned and the values obtained were corrected using regression formulae. Ni concentrations above the accepted hyperaccumulation threshold were found in 20 *Noccaea* species, with the highest value of 42,300 mg kg⁻¹ found in *N. cappadocica* from an ultramafic area in Syria, where hyperaccumulation was found in all scanned specimens. Hyperaccumulation of Zn was detected in 21 taxa within the genus, including all *N. caerulescens* subspecies examined, with 78,000 mg kg⁻¹ as the highest concentration found in *N. caerulescens* subsp. *brachypetala* from Sweden. This taxon, as well as *N. caerulescens* subsp. *caerulescens* and *N. caerulescens* subsp. *virens*, take up both Ni and Zn above the threshold for hyperaccumulation. This phenomenon was also observed in 10 other analyzed taxa of the genus *Noccaea*. However, in *N. cappadocica*, the strongest Ni hyperaccumulator among the taxa studied, the highest Zn concentration was only 41.7 mg kg⁻¹.

KEYWORDS: *elemental profiles, X-ray fluorescence spectroscopy, herbarium specimens, nickel, zinc*

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Ni hyperaccumulation in the flora of Barro Alto, Goiás State, Brazil

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ABSTRACT

The ultramafic soils of Barro Alto, Goiás, Brazil (S14° 58' 6"; W048° 55' 12"), have their origin in serpentinized peridotites. These soils are currently mined as Ni ores. Also, the associations of plants growing on these soils are very different from one soil type to another. The objectives of this work were to better understand the features of Ni hyperaccumulation in the flora of Goiás, especially the relationship between soil type, Ni availability, presence or absence of Ni accumulating species, and levels of Ni in above-ground tissues. The results could support restoration and biodiversity conservation strategies, with a priority target on native species (including Ni-hyperaccumulating plants). The present work was carried out in an area of approximately 10 ha, with native vegetation, at the beginning of Ni ore extraction. According to the topography and vegetation aspect, this area was subdivided into eight subareas, where botanical surveys were carried out and plant and soil samples were collected for chemical analysis. All areas sampled showed characteristics of ultramafic soils, such as pH above 6, low levels of Ca and P, variable levels of K and S, high levels of micronutrients (Cu, Mn, Zn), high Mg/Ca ratio, and shallow and deep soils occur, with the presence of apparent rocks. The bioavailable Ni content in these soils ranged from 100 to 1000 mg kg⁻¹ soil. An initial botanic and soil surveys done in the studied area indicated that differences in the chemical and physical characteristics of the soils influenced the distribution of plant species in the areas and the absorption of nutrients and Ni. During the present botanical survey, more than 1000 plant tissue samples were collected and chemically analyzed. The botanical survey revealed the presence of around 280 species, belonging to 60 botanical families. Among them, 46 species, distributed in 21 families, are considered Ni hyperaccumulators, as they contained > 1000 mg/kg (0.1%) Ni in DW. Although the levels of Ni in the soil can be considered very high, the average levels of Ni accumulated by these species were around 1,500 mg kg⁻¹ DW. The Moraceae and Acanthaceae families had only one (*Dorstenia cayapia*) and two (*Justicia lanstyakii* and *Stenandrium praecox*) species, respectively, that were present in all subareas and consistently hyperaccumulated Ni. The Boraginaceae were represented by a single species (*Euploca salicoides*) also present in all areas, which proved to be a facultative hyperaccumulator of Ni, as the accumulated levels of this element varied greatly among the specimens collected and this was related to the Ni bioavailability in soils. Asteraceae had the highest number of Ni hyperaccumulating species, six in total. Among them, all collected specimens of *Porophyllum obscurum* hyperaccumulated Ni. The Vochysiaceae family is an important component of the Cerrado flora, a savannah-type vegetation that occurs on acidic soils, with high Al saturation and low levels of essential metals. The trait of Al hyperaccumulation is reported to be present in all species of this botanical family (Andrade et al., 2011). On ultramafic soils, however, with high pH and low Al availability, the species *Vochysia rufa* surprisingly did not accumulate Al in the leaf tissues but did Ni. Attention should be given to plants that have adapted to the ecological restrictions imposed by environments rich in metals, such as ultramafic soils, both in strategies for restoring environments impacted by mining and in maintaining areas representative of plant diversity, ensuring of the variability of the genetic pool of genotypes of local origin.

KEYWORDS: *Cerrado region; native species; Ni bioavailability; Ni-hyperaccumulators; recovery of degraded area*

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Performance of Mediterranean hyperaccumulators for Ni agromining in ultramafic soils under humid temperate climate on the NW of Spain

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ABSTRACT

The European economy is highly dependent on imports of Ni, a raw material of high economic importance and agromining is an interesting technology for the recovery of this trace element from low-grade ores such as ultramafic soils. However, climatic conditions, soil properties and metal bioavailability can limit the production of plant biomass and metal accumulation, and consequently the effectiveness of agromining (van der Ent et al., 2015). In this context, one the objectives of the project LIFE-Agromine, funded by the EU, was the establishment of Ni agromining field scale demonstrations on distinct geographical areas and under different edapho-climatic conditions. The present study implemented agromining field trials in an ultramafic soil on the NW of Spain under a humid temperate climate for evaluating the performance of the native Ni hyperaccumulator *Odontarrhena serpyllifolia* and of the Mediterranean hyperaccumulators *Odontarrhena muralis* s.l. and *Bornmuellera emarginata*. The trials also aimed at comparing inorganic (NPK) and organic (cow manure) fertilisation and evaluated 3 cropping seasons. The highest amount of biomass (up to 8 t ha⁻¹) and Ni yield (40 kg ha⁻¹) was obtained with *O. muralis* cultivated with NPK fertilization. Inorganic fertilization was also the best option for the cultivation of *O. serpyllifolia*, although the harvested biomass and Ni yield were much lower than those of *O. muralis* s.l. Conversely, in the case of *B. emarginata* and in the 3 evaluated cropping seasons, the higher Ni yields (up to 11 kg ha⁻¹) were obtained in plots fertilized with cow manure. Interestingly, the harvested biomass of *B. emarginata* was similar in inorganic and organic fertilised plots. The cultivation of the Ni hyperaccumulators for 3 growing seasons did not significantly decrease the Ni availability. Plant ionome and other soil physic chemical parameters were also investigated and are presented.

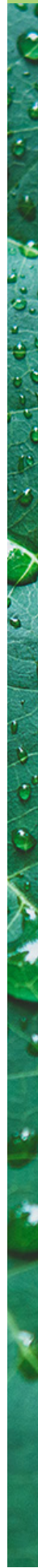
KEYWORDS: *Ni hyperaccumulators, Ni yield, inorganic and organic fertilization, soil Ni availability*

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Session 7 - Plants of other edaphically challenging substrates



Lessons on Ecology and Evolution from the Study of Edaphic Specialization

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ABSTRACT

Rock outcrops and other edaphic islands are characterized by sharp abiotic and biotic gradients, mosaic landscapes, patchiness and isolation, and harsh environments. They offer exceptional opportunities to test macroecological and evolutionary theory, and conservation and restoration practices. Edaphic islands are often distinguished by unique biotic communities with high proportions of rare and endemic species. These communities, often restricted to fragmented islands of harsh substrates, are model settings to investigate the factors and mechanisms contributing to regional and global patterns of diversity and spatial ecology, including metapopulation and metacommunity theory, as well as cross-kingdom interactions. They also provide model organisms for investigating the drivers of adaptive evolution, including the genetic bases for and architecture of traits conferring adaptation and reproductive isolation. Additionally, they offer opportunities to explore cross-adaptation, which is when a trait that evolved for tolerance to one harsh substrate becomes effective as an adaptation for another, allowing species to show cross-tolerance to multiple harsh substrates characterized by a suite of common stressors (i.e., stress resistance syndrome), including habitat bareness, drought, pH, ionic strength, and specific ions. Biota of harsh and nutrient-poor substrates also offer unique challenges for conservation and restoration and are especially prone to stressors associated with climate change, including atmospheric deposition of nitrogen. Much of the research on harsh substrate-biota relations to date has focused on plants of ultramafic and other metal-enriched rock outcrops or saline soils. Recent research on biota of gypsum and other calcareous substrates, solfatara fields, banded iron formations, inselbergs, guano-derived soils, cold deserts, and soils enriched with anthropogenic inputs of nutrients and toxic metals, can help reexamine the biota-harsh soil relationship, especially commonalities and differences across distinct biota-soil type associations, both within and across biomes. Such efforts will also provide opportunities for productive collaboration across research groups with expertise on edaphically distinct communities (e.g., serpentine vs gypsum) or tools of investigation (e.g. ecophysiological vs phylogenomic) relating to key questions on drivers of diversity and community assembly, ecological and evolutionary theory, and conservation and restoration practices.

KEYWORDS: *climate change, co-evolution, cross-kingdom interactions, edaphic endemism, geoeology, harsh environments, rapid evolution*

Current update on floristic diversity of Malagasy inselbergs

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ABSTRACT

Malagasy inselbergs have been identified as one of the three hot spots of inselberg plant diversity with the southeastern Brazil and southwestern Australia. Globally, inselbergs are isolated rock outcrops, mostly consisting of granite or gneiss. They constitute an old terrestrial habitat islands which are widespread throughout the tropics. In Madagascar, they form prominent landscape features, mainly in the central highland regions and have clearly a global importance with regard to their high species richness of certain plant groups such as succulents, terrestrial orchids, resurrection and carnivorous plants. However, most of them are still unexplored and biologically unknown but regular fieldworks conducted each year for more than ten years, on more than 60 Malagasy inselbergs have contributed to a better knowledge about their floristic diversity and habitats. Up to now, more than 1,100 species of vascular plants have been recorded to occur on Malagasy inselbergs, representing ca. 10 % of the Malagasy total flora, comprising ca. 25% of the total genera and ca. 52% of total vascular families present in Madagascar. The most important families as the richest in species are Orchidaceae, Asteraceae, Cyperaceae, Poaceae, Rubiaceae, Fabaceae, Apocynaceae, Lamiaceae, Euphorbiaceae, Asparagaceae, and whereas *Cynorkis*, *Euphorbia*, *Cyperus*, *Aloe*, *Kalanchoe*, *Cynanchum*, *Senecio*, *Exacum*, *Angraecum*, and *Jumellea* represent the most speciose genera. However, Malagasy inselbergs are highly threatened by habitat degradation due to various human pressures (mining, quarrying and fire, the common illegal collection of plants for commercial purposes) resulting in loss of floristic diversity and the ecosystem services they are providing. Ca. third of recorded species have been assessed for their risk of extinction: ca. 20% of them belong to the IUCN Red List threatened categories. Moreover, 147 species are registered in the list of restricted species for trade on CITES Appendices I and II. Inselberg ecosystem in Madagascar requires a special attention for its protection as Inselbergs are poorly represented within the current protected area network.

KEYWORDS: *Malagasy inselbergs, plant diversity, endemics, conservation status, CITES, threats*

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Genomic basis of tolerance to extremely low-pH in an extremophyte (*Carex angustisquama*, Cyperaceae) in highly acidic volcanic soils in Japan

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ABSTRACT

Volcanic acidification creates harsh environment for plants and forces them to cope with various abiotic stresses. Solfatara fields scattered in volcanic areas are among the harshest environments for plants, where rhizotoxicity of extremely low pH (2-3) and high Al³⁺ strongly inhibit plant growth. *Carex angustisquama* is one of the plant specialists in the solfatara fields and has shown to have newly adapted to solfatara fields through speciation. In addition, this species exhibits higher tolerance to low pH stress than the sister species (*C. doenitzii*) growing in non-acidic soils, while there is no difference in Al³⁺ tolerance between them. Therefore, it is needed to identify the genomic basis of low pH tolerance in *C. angustisquama* to understand the evolutionary process of this extremophyte in highly acidic solfatara fields. Here, we have explored the low pH tolerance genes in *C. angustisquama* using comparative genome and transcriptome approaches. Firstly, we obtained Nanopore long reads from *C. angustisquama* (n=33 or 34) to newly construct the draft reference genome consisting of 44 contigs. Then, we performed comparative transcriptome analysis to reveal that several *STOP1* downstream genes, which have shown to play a critical role in *Arabidopsis* low pH tolerance, expressed in response to low pH stress both in *C. angustisquama* and its sister species. On the other hand, highly expressed genes only in *C. angustisquama* included class III peroxidase genes which are involved in cell wall organization. Our comparative genome analysis between *C. angustisquama* and *C. doenitzii* also showed that there was an insertion (~500kb) which harboured significantly high number of class III peroxidase genes than expected from the whole genomic regions. Finally, we tested the contribution of class III peroxidase activities to low pH tolerance in *C. angustisquama* with hydroponic culture experiments using class III peroxidase inhibitor. This pharmaceutical approach showed that the inhibitor caused the decrease in low pH tolerance of *C. angustisquama*. Overall, our results indicated that while *STOP1* downstream genes expressed in response to low pH stress prior to the speciation, class III peroxidase genes played an important role in the adaptation of *C. angustisquama* to highly acidic solfatara fields.

KEYWORDS: *acidic soil, extremophyte, low pH tolerance, volcanic environment,*

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Setting an agenda for gypsum ecosystem research in southern Africa

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ABSTRACT

Gypsum soils occur worldwide and represent natural laboratories of evolution and ecology [1]. The unusual mineral content is a significant barrier to plant growth and yet these soils host highly diverse endemic floras that have evolved independently on five continents. Although gypsum ecosystems are widespread across the African continent, they are poorly understood compared to those of other unusual substrates. Little is known about their conservation status, potential impact of climate change on these systems, and how they respond to mitigation and restoration. A systematic review was conducted to collate research on these topics for Africa, since previous global reviews [1,2] did not cover all biotic diversity and interactions, and had a specific focus which excluded parts of Africa. The current review provides insight into gypsum ecosystem research in Africa, providing a detailed synthesis from approximately 70 papers covering 13 African countries. Research increased from 2000, accounting for half of the papers. Most studies originated from the Horn of Africa (mainly Ethiopia and Somalia), with the majority dealing with gypsum plants and ecosystems of Somalia. Taxonomic research contributed 73% to understanding gypsum floras, followed by vegetation science, phytogeography, community ecology and ecophysiology. Vegetation science contributed extensively to gypsum research in North and Southwestern Africa. Throughout the continent, most research (84%) involved vascular plants, specifically their taxonomy and ecology. The Aizoaceae, Amaranthaceae, Euphorbiaceae, Poaceae and Zygophyllaceae accounted for half of the species mentioned. Southwestern Africa (Namibia and South Africa) had a strong focus on the community ecology of lichen fields. A major shortcoming of the current research on African gypsum ecosystems was that only 27% was conducted by African scientists. Also, research to date was descriptive, did not attempt to understand ecosystem functioning, and had limited continental or cross-continental collaboration in place. The identification of knowledge gaps and prioritisation of research needs for southern Africa during workshops, led to the setting of an agenda for gypsum ecosystem research in the region. To advance the research field in Africa, we need to determine the process, age and rate of gypsum deposition; compile a comprehensive list of species strictly linked to gypsum soils; assess how changes in rainfall patterns influence plant and lichen community composition; characterise and compare plant community composition, diversity, and structure between gypsum and non-gypsum soils; and identify functional traits related to water and nutrient use that impact plant fitness and variation within and among populations.

KEYWORDS: *GYPWORLD, lichens, Namib, plants*

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The Loss and Limitation of Dispersal in Plant Species Restricted to Edaphic Islands in south-east Australia

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ABSTRACT

Sherwin Carlquist was one of the first to document the loss of dispersal in Asteraceae on islands of the Pacific. The isolated nature of islands poses a strong selective force on the dispersal traits in plants simply because large bodies of water act as a significant barrier to long distance dispersal. Therefore, dispersal traits and morphology that favours local dispersal over longer distance dispersal are preferentially selected. Geographically isolated, extreme soils (e.g., serpentine, limestone) might also be seen in the context of ‘edaphic islands’ where we may expect to see a similar loss of dispersal in plant species that are soil specialists. This will likely impact their ability to track their climate niche under increasing temperatures and drought, thereby increasing their extinction risk. We discuss the evidence for the loss of dispersibility in two limestone specialists found in Tertiary and Silurian limestone ‘islands’ from south-east Australia when compared to their widespread counterparts. In particular, we focus on measurements of terminal velocity and model dispersal in a rare Asteraceae and consider elaiosome attractiveness in an ant dispersed Fabaceae.

KEYWORDS: *Limestone, Acacia, Olearia, dispersal*

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Clay Endemism and Affinity in the California Floristic Province

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ABSTRACT

The California Floristic Province of the United States is a geologically and botanically diverse region which includes a substantial number of serpentine-associated and -endemic species¹. While substantial work has been done exploring this component of the flora, no work has yet explored the affinities of plants within the province to clay soils. Clay is an essential component of soil and affects the ability of soils to retain water, ions, and organic matter, all of which are physiologically significant to plant life. As clay content increases, soils exhibit emergent properties which affect plant growth: Soils high in clay exhibit different water dynamics, have higher cation exchange capacity, and require higher quantities of organic matter to stabilize soil aggregates. We compiled a list of species with suspected affinity to clay soils based on local sources^{2,3} for which we sought to quantify clay affinity. We used digitized herbarium records from the California Consortium of Herbaria's CCH2 database for all species and reviewed each species' records to determine the percentage of voucher specimen records for which substrate could be determined, and of those, what percentage indicated a substrate high in clay. Based on the results of these reviews, we found species affinities for clay soils that range from weak association with to strict endemism to clay soils. Past work on serpentine-associated species in the province has used percentage categories for endemism¹ which we have adopted for our work and present as evidence of endemism to high-clay soils in the California Floristic Province. Over 200 species were reviewed for this study, and over 10% of species reviewed as of this abstract's writing are endemic to clay soils, with over 40% showing strong affinity to clay soils.

KEYWORDS: *Geobotany, Edaphic Endemism, Harsh Soils, Clay.*

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Hyperaccumulator *Stanleya pinnata*: In Situ Fitness in Relation to Tissue Selenium Concentration

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ABSTRACT

Earlier studies have shown that *Stanleya pinnata* benefits from selenium hyperaccumulation through ecological benefits and enhanced growth (1,2). So far, there is no evidence for any physiological or ecological constraints. However, no investigation has yet been reported on the effects of Se hyperaccumulation on the overall plant fitness in the field. This research aimed to analyze to what extent the variation in Se accumulation can affect hyperaccumulator *S. pinnata* fitness, as judged from different parameters for physiological and biochemical performance and herbivory while growing in two seasons on two seleniferous sites, Coyote Ridge and Pine Ridge Natural Areas, in Colorado/USA. Natural variation in Se concentration in vegetative and reproductive tissues was determined, and correlations were explored between Se levels with fitness parameters (seed weight, total number of seeds, total number of siliques, total number of leaves), herbivory damage, plant defense compounds (GLS), and total leaf antioxidant capacity. Leaf Se concentration varied between 13- and 55-fold in the two populations, averaging 868 and 2482 mg kg⁻¹ dry weight (DW). Furthermore, 83% and 31% of plants from the two populations showed Se hyperaccumulator levels in leaves (>1000 mg kg⁻¹ DW). In seeds, the Se levels varied 3–4-fold and averaged 3372 and 2267 mg kg⁻¹ DW, well above the hyperaccumulator threshold. Plant size and reproductive parameters were not correlated with Se concentration. There was significant herbivory pressure even on the highest-Se plants, likely from Se-resistant herbivores. We conclude that the variation in Se hyperaccumulation did not appear to enhance or compromise *S. pinnata* fitness in seleniferous habitats within the observed Se range.

KEYWORDS: *selenium; herbivory; glucosinolates; flavonoids; plant fitness; seleniferous soil*

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Lessons Learned and Paths Forward: Ensuring Long-Term Success of Revegetation Efforts on Metal-Contaminated Mine Tailings in Arid and Semi-Arid Ecosystems

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ABSTRACT

Phytoremediation is a sustainable and eco-friendly approach to remediate mining legacy sites. However, unfavorable biogeochemical characteristics of mine tailing deposits, including low fertility and high salinity, often limit the success of phytoremediation, particularly in arid and semi-arid areas. Previous phytoremediation efforts at the highly contaminated Iron King–Humboldt Smelter (IKHS) Superfund Site, located in Dewey-Humboldt, AZ, USA involved adding compost in varying proportions to the contaminated soil to promote plant growth. The site is contaminated with arsenic, lead, zinc, and iron, with an average concentration of ~4000 mg kg⁻¹, 2400 mg kg⁻¹, 6100 mg kg⁻¹ and 106000 mg kg⁻¹, respectively [1]. Currently, 13 years after compost amendment, only a few species, including *Atriplex lentiformis*, had managed to survive in this hostile environment. To further investigate the potential of the IKHS tailings material as a substrate for successful revegetation, we assessed the current state of the soil microbial community diversity and structure at this historically revegetated site and evaluated the effect of a second compost amendment on plant-microbe interactions. Under controlled conditions, we monitored temporal changes in plant performance and root systems across eight different metal-contamination levels of mine tailings. We observed successful plant development, suggesting the biological activity of microorganisms added to tailings a long time ago. These findings will facilitate the development of effective site-tailored strategies for the sustainable phytoremediation of metal-contaminated and nutrient-poor mine tailings.

KEYWORDS: *mine tailings; revegetation; phytoremediation; metal tolerance; soil microbiome*

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The Vertic Clay Flora of the San Joaquin Desert: Niche, Competition & Floral Mosaics on a Novel Substrate

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ABSTRACT

Plant communities that occur on edaphically heterogeneous landscapes are ideal systems for exploring questions of niche and community assembly. Species with affinity for harsh substrates often have well-defined edaphic niches and are ideal models for testing ecological and evolutionary theory. While plants on substrates such as serpentine and gypsum have received much attention, those on other harsh substrates, including on vertic clay soils, have gone unstudied. Vertic clay soils are both chemically and physically challenging to plant establishment and productivity. Plant communities associated with vertic clay soils of the San Joaquin Desert appear to have a distinctive mosaic pattern of species distribution that reflect differences in soil properties across the landscape. We conducted fieldwork and a pot study with 12 native annual plant species with an affinity for vertic clay soils to determine whether the vertic clay soils at two sites in the San Joaquin Desert were heterogeneous, whether soil heterogeneity determined the pattern of species distributions observed, and to examine the competition effects of an invasive annual grass (*Bromus madritensis*) on these species. We found that the vertic clay soils at both of our sites are internally heterogeneous, that soil heterogeneity does appear to shape the patchy distribution of species at both these sites, and that these species have different realized edaphic niches. We utilized treatment soils spanning a gradient of chemical stress present at both study sites in our pot study and found that competition from *B. madritensis* reduces biomass for all species, and that the effect of competition differed between soil types. Further, we found that species' edaphic niche optima shift when competition is present, and that competitive ability differed across the gradient of edaphic stress in our treatment soils.

KEYWORDS: *Community Ecology, Edaphic Niche, Vertic Clay, Clay, Invasive Annual Grass, Desert Communities.*

Violets of the Allchar arsenic-thallium deposit (Republic of North Macedonia) – proof of hyperaccumulation

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ABSTRACT

The abandoned Allchar mine in North Macedonia is one of the largest thallium deposits in the world and the area with the strongest known grades of As and Tl mineralization. The highly unusual mineral composition of the soil, considered to be one of the most toxic in the world, hosts a unique flora with a high proportion of endemic species. Among the 7 endemic taxa discovered so far in the Allchar area, there are two *Viola* species, *V. allchariensis* and *V. arsenica*. Considering that they belong to the section *Melanium*, which includes a large number of metallophytes, these taxa, along with *V. tricolor* subsp. *macedonica* (which also grows in Allchar) were studied for their potential to accumulate metal(loid)s. Although the species differed greatly in terms of their metal(loid) accumulation characteristics, the analysis revealed Tl hyperaccumulation in all three species, whereas As concentrations were highly variable. To exclude surficial contamination with soil particles as a reason for the high prevailing foliar Tl and As concentrations, dehydrated shoots were analyzed by X-ray fluorescence microscopy at beamline P06 of PETRA III (Deutsches Elektronen-Synchrotron, DESY). The differences in the distribution of Tl compared to other elements indicate its endogenous origin and hyperaccumulating character of the species analyzed. In *V. arsenica*, As also has an endogenous origin. However, the high As concentrations in *V. tricolor* subsp. *macedonica* and *V. allchariensis* are most likely due to soil particle contamination, as the hotspots correspond to those of other common soil elements.

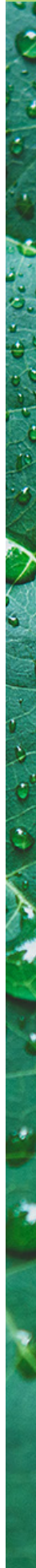
KEYWORDS: *hyperaccumulator, synchrotron-based X-ray fluorescence microscopy, elemental distribution*

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Session 8 - Agromining & phytotechnologies



Agromining in Sabah, Malaysia: Current Status of Knowledge and Future Potentials

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ABSTRACT

Agromining is a novel agricultural technology in which hyperaccumulator plants are grown and their biomass harvested every 6 months. Also known as “Metal Farming” where harvested leaves are then processed to extract nickel metal. Hyperaccumulators are plants that can naturally accumulate up to 3% of nickel in their leaves and only grow on ultramafic soil. As Sabah has the largest ultramafic land in Malaysia, almost 5% of its total land area, it has become one of the most important places in the world for hyperaccumulators with over 30 species discovered. To date, exploration is still on going with a research collaboration with a local university, Universiti Teknologi MARA (UiTM) and reserve manager, Sabah Parks. Recently, studies have been carried out to verify the hyperaccumulating capabilities and capacities of hypernickelophores, such as *Phyllanthus rufuschaneyi* in pot experiments and trial plots in Kg Pahu, Ranau. Currently, a pilot study has commenced to carry out breeding program at Kg Turuntungan, Ranau by Econick Sabah Sdn. Bhd. It is projected that in 5 to 6 years, Econick Sabah Sdn Bhd will be able to create a local agricultural industry based on novel agromining that will improve the quality and quantity of farming activities in Sabah. Eventually, this will create local employment besides generating economic opportunities for the villages in the Nalumad-Takutan-Pinawantai-Turuntungan-Pahu-Monggis area through agromining contract farming.

KEYWORDS: *hyperaccumulators, serpentine, metal crop, Phyllanthus*

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Developing speed-breeding techniques for European nickel metal crops

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ABSTRACT

The breeding of new cultivars of most domesticated crop plants is a process that usually takes decades if not centuries. Typically, four to six generations of inbreeding are required to develop genetically stable homozygous parent lines for evaluation of agronomic traits [1]. This process is therefore very time-consuming for field grown crops where the maximum generation cycle per year is limited to only one or two cycles [2]. In order to accelerate the plant breeding and selection programmes an innovative methods of plant cultivation ('speed breeding') has been developed in recent years with the aim of reducing the seed-to-seed generation time. The leading principles of speed breeding systems are the cultivation of plants under long or continuous light (22–24 hrs) followed by the harvesting and germination of immature seed [1]. Speed breeding protocols have been successfully developed for several species, including spring wheat, barley, canola, and chickpea [3]. In these scenarios generations per year increases from two/three in the glasshouse (with no heating or additional lightning), to over six per years with speed breeding systems. The development of speed breeding has thus far been limited to common (food) crop plants, but could be applied to other plant species of commercial interest, such as nickel metal crops (*i.e.*, hyperaccumulator plants) used in agromining. Furthermore, these species can especially benefit from this technique given their biennial-perennial habit under natural conditions. However, speed breeding methodologies have not yet been developed for these species (or indeed for any hyperaccumulator species), despite the crucial importance of selection and breeding programmes aimed at improving their nickel yield in large-scale commercial agromining operations. The overall aim of this study is to develop a robust and effective speed breeding system that can reliably generate at least two, but ideally four, seed-to-seed generations of the key metal crops *Odontarrhena chalcidica*, *Bornmuellera emarginata* and *Bornmuellera tymphaea* per year. At present the project is in the initial phase, and since late 2022 we have been collecting phenological data on the three selected hyperaccumulator species to identify the optimum cultivation method using soil, hydroponics, or aeroponics cultivation. Furthermore, we are identifying the best growing conditions for light requirements (light spectrum and duration), temperature (during growth, vernalization, and flowering) and nutrient supply that will accelerate plant development to flowering and seed set. The data collection will be completed in April–May 2023 and the preliminary findings will be presented at the ICSE in June.

KEYWORDS: *Ni-agromining, hyperaccumulator cultivation techniques, hydroponics, aeroponics, plant phenology*

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Environmental assessment tools applied to the agromining chain, from the field to marketable products

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ABSTRACT

In the long term, the cultivation of hyperaccumulating plants for the production of metals makes it possible to supplement (or to substitute) a part of the conventional mining production. This phytotechnology has the advantage to enable the exploitation of resources with low metal concentrations (*e.g.* mine tailings), or with recalcitrant deposits. But, one of the main advantages of this nature-based solution is the positive image, given by the use of plants (natural process, landscape conservation *etc.*). However, this way of metal recovery is not without any damage to the environment (*e.g.* consumption of chemicals, energy) and it is necessary to evaluate the impacts of agromining chains. The aim of the work is to apply some of environmental analysis tools and to assess the relevance in agromining contexts. Two different tools have been studied: (i) the E-factor, coming from the green chemistry, corresponding to the mass of waste produced per kilo of desired product, tested on a laboratory scale for the production of rare earth carbonate from the fern *Dicranopteris linearis*, and (ii) life cycle assessment (LCA), on a pilot scale, for the production of nickel salts from *Odontarrhena chalcidica*, grown in Albania. In the first case, the results show that the E-factor is relevant for the selection of atom-efficient processes, very early in the chemical design, and for their optimization. Thus, for the production of rare earth carbonate, it was reduced by a factor of almost 4 (from 732 to 197 kg/kg), by changing the chemical approach. This method is fast and efficient, but is very piecemeal: for instance, it does not consider energy aspects. In the second case, it could be shown that a large part of the environmental impacts is related to plant cultivation. Nickel agromining in serpentine soils is environmentally positive, if energy is recovered during ash production. Moreover, it limits the transfer of heavy compounds into the food chain. LCA is a much more global analysis, indicating hot spots in the whole chain and taking into account pollution transfers. However, it is only reliable when the technology is robust and already implemented on a large scale. Therefore, the development of the agromining chain can be accompanied by various environmental analysis tools, from the beginning of the concept to the semi-industrial technology readiness level. This exercise is mandatory for agromining to remain a high environmental value process.

KEYWORDS: *Agromining, environmental assessment, nickel, rare earths, life cycle assessment, E-factor*

Nickel accumulation and spatial distribution of metals assessed by SR- μ XRF in tropical hyperaccumulators

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ABSTRACT

The relatively small number of promising hyperaccumulators in South America has hindered nickel (Ni) agromining in this part of the world. For instance, most of the Brazilian hyperaccumulators have low biomass and accumulate < 1% Ni in shoots. Besides, even for the known hyperaccumulators in Brazil, the potential to concentrate and distribute Ni in different plant tissues and the mechanisms they use to deal with high Ni concentrations still need to be understood. Such knowledge is fundamental to selecting the most promising species to make agromining technically and economically feasible in tropical settings. Here, we aimed to evaluate the potential for Ni accumulation in the Brazilian hyperaccumulators *Pfaffia sarcophylla*, *Justicia lanstykii*, and *Lippia lupulina* growing naturally in an ultramafic soil in Niquelândia, Goiás State. In addition, species from South Africa (*Berkheya coddii*) and Mexico (*Blepharidium guatemalense*) that can adapt to Brazil were grown in pots containing ultramafic soil. The Ni concentration in the leaves of the five species was determined by handheld x-ray fluorescence (pXRF). Furthermore, synchrotron X-ray microfluorescence evaluated the spatial distribution of metals in whole leaves. We also performed histological sections and histochemical tests on the most promising species to visualize Ni distribution at the cellular level. The results showed that Ni concentrations in the plants varied from 2222.0 (*P. sarcophylla*) to 14960.0 mg kg⁻¹ (*B. guatemalense*). The other species accumulated 3042.5 (*L. lupulina*), 3863.0 (*J. lanstykii*) and 11387.0 (*B. coddii*) mg kg⁻¹ of Ni in the shoots. Nickel, Mn, and Co spatial distribution in the leaf blade was similar in all species, with preferential accumulation in the leaf margins and veins. Microscopic analysis in *B. guatemalense* showed that Ni was concentrated in the epidermis and phloem, suggesting translocation to younger tissues and likely defense mechanism against herbivores. Our data suggest a low potential of the Brazilian hyperaccumulators tested here for commercial agromining. However, *B. guatemalense* and *B. coddii* high Ni accumulation makes these species candidates for tropical agromining. Field trials testing these species are to be initiated in ultramafic sites of Northeastern Brazil.

KEYWORDS: agromining; metallophytes; phytomining.

Optimal co-cropping densities of *Noccaea caerulescens* with *Sedum plumbizincicola* for zinc *in situ* phytoextraction depend on metal bioavailability

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ABSTRACT

In the coming years, co-cropping hyperaccumulator plants could emerge as a promising agricultural practice that mitigates trace metal contamination of soils while addressing the challenges of climate change and biodiversity loss. But currently, multi-species covers of zinc (Zn) and cadmium (Cd) hyperaccumulating plants have never been tested under field conditions. This study designed the first-ever field trials conducted between *Noccaea caerulescens* Ganges ecotype (NcG) and *Sedum plumbizincicola* (S), two hyperaccumulators from different botanical families that have been extensively described as laboratory model species. These two hyperaccumulators may show complementary traits of interest for Zn/Cd phytoextraction under *in situ* conditions. Our objectives were to identify optimal co-cropping conditions of the NcG-S association, in terms of plant density, to improve plant development and Zn/Cd extraction efficiency. For that purpose, two synchronous 50 m² field experiments were implemented over five months with identical soil cover systems and with the same intercropping densities, tested in triplicate: (i) 11 NcG plants m⁻² - 13 S plants m⁻² (density 1 designated D1), (ii) 21 NcG plants m⁻² - 11 S plants m⁻² (D2), (iii) 32 NcG plants m⁻² - 8 S plants m⁻² (D3). Soil A belonged to an industrial wasteland amended with compost characterized by subalkaline pH_{water} (7.8 ± 0.1), while soil B was an agricultural land with neutral pH_{water} (7.1 ± 0.2), both not limiting in terms of soil fertility. However, the soil Zn resources in these two substrates differed greatly regarding DTPA-extractable levels (37 ± 12 mg kg⁻¹ in soil A compared to 15 ± 4 mg kg⁻¹ in soil B) and pseudo-total concentrations (237 ± 48 mg kg⁻¹ in soil A and 87 ± 17 mg kg⁻¹ in soil B). Cd pseudo-total concentration reached 4.2 ± 4.2 (1.1 - 13.6) mg kg⁻¹ in soil A but was limited to 0.27 ± 0.03 mg kg⁻¹ in soil B. DTPA-extractable Cd levels were below 0.06 mg kg⁻¹ in soil A and amounted to 0.10 ± 0.01 mg kg⁻¹ in soil B. Unexpectedly, contrasting responses of co-cropping systems were reported between the two soils. In soil A, greater Zn phytoextraction efficiency was achieved at D1 (+33%, compared to D2 and D3), mainly due to a significant increase in S-shoot biomass whereas shoot Zn concentrations differed only slightly between the species and densities tested. With more limiting Zn resources, the opposite pattern was found in soil B: the phytoextraction of Zn was considerably higher at D3 compared to D1 (+73%), mainly driven by a net increase in NcG shoot biomass with density. This effect was also accompanied by a significant decrease (-50%) in Zn accumulation of S-shoots compared to NcG, which maintained the same level of performance in both soils. Surprisingly, *S. plumbizincicola* did not accumulate Cd in shoots (< 4 mg kg⁻¹) under these edaphic conditions, unlike *N. caerulescens* which was able to (hyper)accumulate it *in situ*.

KEYWORDS: agromining, field trials, intercropping, hyperaccumulator plants, zinc, cadmium

Biotechnological tools for improving Ni agromining in ultramafic soils

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ABSTRACT

Agromining cultivates Ni-hyperaccumulator plants for recovering valuable Ni-products from the harvested biomass. Agromining of ultramafic agricultural soils is considered an eco-friendly option for the sustainable management and re-valorisation of these soils. The optimization of plant growth and Ni extraction is of great importance for the cost-effective implementation of agromining. In this regard, the use of plant growth regulators (PGRs) or phytohormones (Cabello-Conejo *et al.* 2014) or bioinoculants (Becerra-Castro *et al.* 2013) has been proposed as an effective means of maximising Ni yields. The study presented established agromining field experiments with the Ni hyperaccumulating species *Odontarrhena muralis*, *O. serpyllifolia* and *Bornmuellera emarginata* in an ultramafic soil on NW of Spain and tested the application of the phytohormones Promalin® and Kelpak® for improving the harvested Ni enriched plant biomass and Ni yield. In addition, the effect of the bioinoculation of *Odontarrhena muralis* with 3 different plant growth promoting rhizobacteria (*Paenarthrobacter nitroguajacolicus* strain LA44, *Pseudoarthrobacter oxydans* strain SBA82 and *Variovorax paradoxus* strain AB30) was also evaluated. The treatment with of Promalin® improved the biomass and Ni yield obtained with *B. emarginata*; however, did not promote the growth of the other hyperaccumulators. Conversely, Kelpak tended to decrease plant growth. Regarding the bioinocula, the strains LA44 and SBA82 significantly improved the biomass and Ni yield obtained with *O. muralis*. The effect of the treatments evaluated on plant ionome and on soil physic-chemical analysis, including Ni availability, were also analysed and will be presented.

KEYWORDS: *Ni agromining, phytohormones, bioinocula, plant growth promotion*

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Improving plant growth on mine tailings by using N-rich residual materials from mine water treatment

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ABSTRACT

Ammonia treatment in contaminated mine water generates N-rich residual materials such as moving bed biofilm reactor (MBBR) biomass and N-rich zeolites (biological and geochemical treatment processes, respectively) that could be valorised as slow-release source of nitrogen for establishing plants on mine tailings. The objective of this study was to evaluate comparatively, in a 3-months pot experiment, the effect of mineral NPK fertilizer, MBBR biomass, and N-rich zeolites (at a rate of 100 kg total N/ha) on above- and below-ground growth and foliar nutrient concentrations of a Fabaceae and several Poaceae species on non-acid-generating gold mine tailings with low initial concentrations of mobile trace metals. Since N-rich zeolite and MBBR biomass obtained from gold mine effluent treatment contain salinity and trace metals, potential phytoaccumulation of elements in leaves was also investigated. N-rich zeolite addition to tailings did not improve the total, above and belowground biomass production of the tested plants compared to tailings alone, despite greater foliar N concentrations in *Melilotus albus* and some tested Poaceae. On the contrary, MBBR biomass or NPK fertilizer addition induced similar biomass and foliar N increase of plants that were greater than on tailings alone, except for *M. albus*. In Poaceae leaves, Se concentrations were up to 5.9 mg/kg in MBBR biomass treatment, while zeolite treatments entailed Na concentrations up to 11 g/kg, and Mn concentrations increased in both (up to 331 mg/kg) compared to control treatments. Some foliar samples presented high concentrations of Cr, especially in MBBR treatment. Based on the findings, MBBR biomass could be an interesting alternative to NPK fertilizer to phytostabilize the tested mine tailings. This study did not show that N-rich zeolite could be used to improve plant biomass production on the tested tailings, but this result could be different in tailings with greater mobile trace metal concentrations since zeolites are known to help remediate contaminated soils via sorption of cationic species.

KEYWORDS: Agronomic grasses and legumes, slow-release N fertilization, zeolite, MBBR biomass, gold mine tailings revegetation

LIFE-AGROMINE, a pilot-scale demonstration carried out at EU level

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ABSTRACT

The objective of LIFE-AGROMINE was to demonstrate the feasibility of the agromining chain at pilot-scale, from cropping nickel-hyperaccumulators to the recovery of bioenergy and valuable Ni-rich products. This includes phytomining on natural Ni-enriched agricultural ultramafic soils to degraded lands and industrial waste materials. The project was also designed to provide a complete life cycle assessment (LCA) of the full chain along with a techno-economic study that would help to select the best technological ways to implement agromining and thus orientate the future development of the profitable options. Finally, dissemination of best practices identified during the project was carried out with the participation of SMEs wanting to develop the business (business & technical meetings, training workshops). Social acceptability of agromining through intense communication to public was also targeted. The results obtained showed very encouraging technical perspectives [1] [2] and the extremely positive impact on stakeholders will ensure with no doubt the success of LIFE-AGROMINE for both its level of achievement compared to the initial goals as both TRL 7 was reached, dissemination towards all targets was successful (i.e. the attention of law-makers was brought in an efficient way to the regulation aspects needed) and the market uptake of this low-C and environmentally-friendly technology at European level and the perspectives for business development are serious. “After-LIFE”, ECONICK will scale up the technology to industrial pilot-scale with the help of industrial partners in collaboration with the network of beneficiaries to target the sustainable sourcing of nickel for the stainless steel and the glass industries.

KEYWORDS: *phytomining, nickel, hyperaccumulators, nature-based solution, circular economy, stakeholders*

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Preliminary results of optimizing Ni phytoextraction by *Odontarrhena lesbiaca*

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ABSTRACT

A pot experiment was conducted for nine months in environmental conditions to test the potential effect of agronomic practices such as soil amendment, N fertilization, and varying plant density on Ni phytoextraction yield of *Odontarrhena lesbiaca*, a metallophyte endemic to the serpentine soils of Lesbos [1]. Four treatments were applied: fertilization with mineral N fertilizer; co-cropping for three months with the legume *Trifolium angustifolium*; fertilization with compost, and control [2, 3]. For each treatment, plant density of 1, 2 and 4 plants per pot was tested. Each combination of treatment and density was run in six replicates. In all cases, a Ni-rich soil from an adjacent ultramafic locality was used, amended before sowing with granulated CaCO₃ so as to improve the Mg/Ca ratio and with KH₂PO₄ to adjust the P and K contents. Before harvesting a number of growth parameters and functional traits were measured in the plants whereas upon harvesting total dry leaf and stem biomass was measured. Elemental concentrations in stems and leaves were determined using ICP-OES after acid mineralization (HNO₃ and H₂O₂) and Ni phytoextracted mass was calculated. Plant growth was mostly controlled by density and single plants per pot were higher, had a greater number of longer and wider branches and thicker but shorter leaves. Only the width of branches was affected by the treatment; the widest branches were measured in plants treated with mineral fertilizer and the narrowest in those co-cropped with legume. On the other hand, total dry biomass production per pot was determined only by the application of mineral fertilizer, which resulted in a ca. 20% increase in yield as compared to the other treatments. As for the functional traits, both the fertilization regime and density played a role in their variability, but not in a consistent way. The supply of nitrogen did not favour Ni concentration in the aboveground biomass of the plants, which ranged from 2.3 to 10.0 g kg⁻¹ (mean ± sd: 5.6 ± 1.5 g kg⁻¹). However, Ni concentration varied with density in the order 2 ≥ 1 ≥ 4. Despite the differences in plant biomass production and Ni content, Ni phytoextracted mass was similar in all cases, 99 ± 30 mg Ni/pot (range: 40 – 190 mg Ni/pot). This finding suggests that further investigation is needed in order to optimize Ni yield and to enhance *O. lesbiaca*'s potential for phytomining, in terms of type and dosage of fertilizer, as well as length of experimental observation, given the perennial nature of the species.

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KEYWORDS: *Alyssum lesbiacum*; phytomining; legume co-cropping; fertilization; compost

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Changes on soil bacterial communities after an agromining field trial in the NW of the Iberian Peninsula

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ABSTRACT

Most of field assessments for agromining potential demonstrations in Europe have mainly been carried out in Mediterranean ultramafic regions (Bani et al., 2015a; Bani et al., 2015b). Assessing the performance of agromining crops in other geographical areas and under different climatic conditions are necessary for the optimization of the process and determination of its viability and/or effects on ecosystem services. The aim of this work was to evaluate changes on soil bacterial communities within an agromining system during a field experiment in an ultramafic outcrop under a humid-temperate climate in NW Spain. On one side, we evaluated, using Illumina Miseq barcoding sequencing, the soil bacterial communities in monoculture plots 1 month ($t = 0$) after planting *Odontarrhena muralis*, *O. serpyllifolia* and *Bornmuellera emarginata* for the first time as well as during the second cropping season of agromining in the same plots. The bacterial communities were also compared with those associated to native plants of *O. serpyllifolia* spontaneously growing at the same ultramafic area. On the other hand, soil bacterial communities in fertilised plots (NPK fertilisation and manure amendment) planted with *O. muralis* and *B. emarginata* were also studied. Firstly, a clustering analysis was carried out for the communities in the monoculture plots: two clusters including samples from the two different sampling times were obtained, differentiating bacterial communities at the initial phase of the agromining from those present in soil after 2 cropping seasons. Moreover, this second cluster was located closer to native plants of *O. serpyllifolia*, which suggest a progression of bacterial communities approaching those characteristics of native plants. The community was dominated by Actinobacteria and Proteobacteria, and the third most abundant phylum was Bacteroidetes, while the three most abundant genera were *Nocardioides*, *Blastococcus* and *Bradyrhizobium*. Some changes in genera abundance after two cropping seasons were observed, approaching the values found in the native plants: the abundance of *Blastococcus* increased with time, while the abundance of *Sphingomonas* and *Massilia* decreased. In the case of the fertilised plots, two main cluster were obtained separating communities in soils receiving NPK from those fertilised with manure. Interestingly, planted soils clustered separately from non-planted only in the case of manure fertilisation. As in the case of the monoculture, community was dominated by Actinobacteria and Proteobacteria, however, the third most abundant phylum was Acidobacteria. Finally, correlation analysis between abundant OTUs and soil and plant properties showed a higher number of positive correlations in the NPK treatment. Moreover, some OTUs, belonging mainly to the genera *Mycobacterium*, *Nocardioides* and *Geodermatophilus* presenting the highest number of positive correlations and will be further studied.

KEYWORDS: Agromining, soil bacteria, agronomic practices

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Substrate formulation for agromining of cobalt from aged mine tailings

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ABSTRACT

Secondary resources are potential substrates for alternative mining strategies, such as agromining. The increasing demand of strategic and critical metals (SCM), such as Co and Ni, which are key elements for the energy transition, have encouraged their recovery from secondary resources. Agromining is the chain of processes where hyperaccumulating plants are grown to extract SCM from various substrates and recover these elements with a combination of pyro/hydrometallurgy. Repurposing of mine tailings through agromining seems to be a promising approach for both economical and environmental reasons. Implementation of agromining requires adapted substrates for plant growth, and development as well as subsequent uptake and concentration of elements in the plant tissues. However, growing plants on mine tailings can be challenging due to several factors (*e.g.*, high contents of contaminants, important lack of nutrients and poor soil structure). Therefore, the present study aims to identify the most favorable conditions for the growth of hyperaccumulator plant species and to quantify their potential to accumulate Co and Ni from aged mine tailings. The selected mine tailing materials contained 1310 mg kg⁻¹ Co, 943 mg kg⁻¹ Ni and 5245 mg kg⁻¹ As. The viability of the selected plant seeds (*O. chalcidica*, *B. emarginata*, *S. canadensis*, *P. arundinacea* and *L. perenne*) was tested through germination tests, and dormancy was lifted in some species through cold stratification. Some of these species were selected based on their demonstrated ability to hyperaccumulate Ni or Co, while others were chosen for their accumulation potential and ability to grow in extreme environments, for example on degraded lands (spontaneously colonized the mine tailings site). An organic amendment (table compost) was selected to improve tailings fertility, structure, and properties to create favorable physical and chemical properties for plant growth and development. The results showed that germination rates varied from 0 to 83% depending on the species and the type of substrate used. The germination rates of *O. chalcidica* on aged mine tailings varied depending on the proportion of amendment to residues (0/100 to 70/30), with values ranging from 0 to 40%. The concentrations of metallic elements in plants will be determined to evaluate the accumulation potential of Co and Ni on such substrate. While growing plants on mine tailings represents a significant challenge, identifying the most favorable conditions for the growth and accumulation of hyperaccumulator plant species has the potential to unlock the valuable resources in secondary sources through agromining.

KEYWORDS: *agromining, mine tailings repurposing, phytoextraction, strategic and critical metals*

Assisting nickel agromining using sustainable amendments

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ABSTRACT

Agromining is dedicated to the rehabilitation of ultramafic soils but the success of phytoextraction by hyperaccumulating plants depends on different parameters such as plant tolerance to metallic stress, high biomass production and a high concentration of metals transferred from the roots to the aerial parts of the hyperaccumulators [1]. During the last decade, many studies have focused on optimizing of metal phytoextraction, using chemical methods and chelating agents [2]. But, as for conventional agriculture, it is important to replace pesticides and mineral fertilizers with eco-efficient strategies, because of resource depletion and the increasing global demand for mineral fertilizers, in addition of the serious threat to human health and the environment that pose agro-chemicals. The aims of this study were to test, under controlled conditions, the interest of using 4 sustainable solutions dedicated to the optimization of nickel phytoextraction by *Odontarrhena chalcidica*: a biostimulant (humic acid and fulvic acid), a combination of a biostimulant and plant defence stimulator product composed of algae extracts, artificial root exudates, and a biodegradable metal chelator (EDDS). Their effects on Ni phytoextraction capacity, on growth and physiology of *O. chalcidica*, but also on the physicochemical soil characteristics were studied in comparison with a conventional fertilizer (ammonium nitrate). We also studied their effects on the richness and diversity of rhizosphere and endosphere bacteria. Biostimulants appeared to be potentially interesting for improving nickel phytoextraction and plant biomass production and showed a positive effect on bacterial richness and diversity. Conversely, artificial exudates and ammonium nitrate appeared to be of little interest for the optimisation of nickel agromining.

KEYWORDS: agromining, nickel hyperaccumulator, sustainable amendments

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Citric acid-assisted accumulation of nickel by *Blepharidium guatemalense*

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ABSTRACT

The success of Ni agromining depends mainly on the metal availability in the soil or substrate and its accumulation on the plant's aerial biomass. Techniques that increase the Ni availability in the soil and the Ni plant uptake can improve agromining performance. For example, natural chelating agents such as citric acid can complex Ni and increase its concentration in the soil solution. Induced-phytoextraction for Ni using citric acid has been scarcely tested in Ni hyperaccumulators cultivated in ultramafic soils, with conflicting results. Here, we aimed to assess the effect of citric acid on Ni availability in the soil and accumulation by the hyperaccumulator *Blepharidium guatemalense*. Plants were grown for 90 days in pots containing ultramafic soil from Buenos Aires, Pernambuco state, Brazil. The citric acid (40 mmol kg⁻¹ soil) was applied to the soil on the 85th day of cultivation. The experiment was carried out in a completely randomized design with 4 replications. Our results showed that applying citric acid doubled the soil Ni availability. The Ni content in the aerial biomass of *B. guatemalense* was 14960 mg kg⁻¹, with 18% of Ni in the bio-ore. Plants treated with citric acid showed an increase in the translocation of Ni from roots to shoots and, consequently, in the Ni concentration in the shoots (18405 mg kg⁻¹); in turn, the Ni content in the bio-ore reached 30%. Our results need to be confirmed under field conditions but indicate that citric acid can increase the efficiency of Ni agromining with *B. guatemalense* in ultramafic soils.

KEYWORDS: *bio-ore, chelation, heavy metal, hyperaccumulator, phytomining.*

Cutting Propagation of *Bornmuellera tymphaea*, a Nickel Hyperaccumulator for Use in Agromining: Effects of Substrate and Auxins on Stem Cuttings

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ABSTRACT

Nickel hyperaccumulator *Bornmuellera tymphaea* has been evaluated for use in agromining in Greece with very promising results. This study aims to verify the possibility of propagating this species through cutting-based vegetative propagation, provides rapid multiplication and clonal propagation of genotypes having a high level of genetic traits and offers obvious advantages over propagation by seed. Apical cuttings, 10 cm long, obtained from plants from the field, were propagated in benches under misting at greenhouse conditions. Four treatments based on the substrate were evaluated: a) perlite, b) peat + perlite 1:1 (v/v), c) perlite + ultramafic soil 1:1 (v/v) and d) ultramafic soil. Completely randomized blocks were used in a 4x2 factorial design, using four different propagation substrates and two different levels of auxins, with three replicates of 70 cuttings. The response of perlite medium was best in terms of rooting 75 % without IBA and 71,33% with IBA, the dry weight of roots was 0,18 g and of the shoots 0,34 g. Peat + perlite 1:1 (v/v) medium also resulted in rooting in more than 55% of cuttings whereas cuttings raised in ultramafic soil showed very low rooting (<27%). Based on the findings, *B. tymphaea* has an aptitude for vegetative propagation by cuttings with survival values and rooting greater than 70%. IBA presence did not have beneficial impact on the cuttings rooting percentage.

KEYWORDS: vegetative propagation, ultramafic soil, perlite, peat, IBA

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Exploring the Potential of Nickel Agromining in Western Macedonia, Greece: An Analysis of Land Use and Geological Factors.

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ABSTRACT

The global demand for nickel is increasing and it is considered a product of high economic importance. In Western Macedonia, Greece, there are significant opportunities for nickel agromining due to the presence of large areas of ultramafic soils, which are rich in nickel but not very productive for conventional farming. The aim of this work is to explore the possibility of nickel agromining in the Western Macedonia region. The study combines information from various sources including bibliographic data on agromining, experimental field applications under the LIFE Agromine project, land uses, and geological data in the region. The information is processed in GIS software to identify the areas most suitable for agromining. Results show that there are a total of 14.235 hectares in Western Macedonia that are potentially suitable for agromining applications, and the study provides maps outlining these areas. These findings demonstrate the significant potential for nickel agromining in the Western Macedonia region and support the implementation of pilot applications for further performance assessment, contributing to the growing demand for nickel globally.

KEYWORDS: *nickel, ultramafic, GIS, geological data*

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Minimizing the environmental impact of agromining by membrane processes

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ABSTRACT

Agromining is the recovery of valuable heavy metals from soils with high metal concentrations, using hyperaccumulator plants, to produce value added products [1]. It has been successfully applied to serpentine soil in Albania, Greece and Malaysia, achieving a nickel yield of up to 100kg nickel per hectare [2]. However, nickel recovery from plants ashes by hydrometallurgy requires a leaching step, using a very large quantity of reagent, mainly sulphuric acid. It is important to note that only 30% of the acid used is consumed, the rest is mandatory but does not react and need to be neutralized then. Proton recycling could be a good option to decrease the environmental impact of this kind of processes. Therefore, the aim of this presentation is to present the approach and first results of a proton recycling step from an hyperaccumulator (*Odontarrhena chalcidica*) ash leachate, using a nanofiltration membrane. This is now possible, thanks to the emergence of new membranes on the market, resisting to very low pH and high ionic strengths (Duracid NF, a Suez brand membrane, in our case). First results show that the ionic strength of the leachate has a great influence on the retention of the different elements. Leachate dilution by a factor of four increased the retention of nickel from 30% to over 80%. This phenomenon is could be explained by a change in the effective pore size (pore swelling) due to accumulation of counterions inside pores (3). A good compromise must therefore be found between a discharge rate, which is desired to be as high as possible to ensure the lowest possible concentration of metals in the permeate, and a volume of effluent, desired to be as low as possible. With this process, it is technologically possible to recycle up to 2/3 of the acid used in the leaching process. Modelling the mass transfer through the membrane as a function of leachate element speciation is in progress. This work will allow the simulation of proton recovery potential. Coupled with a life-cycle assessment, as well as a technical-economic analysis, it will allow the best design of agromining process. Moreover, this approach may be deployed to other hydrometallurgical leachates.

KEYWORDS: Agromining, Nanofiltration, Nickel

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Performance of Mediterranean hyperaccumulators for Ni agromining in ultramafic soils under humid temperate climate on the NW of Spain

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ABSTRACT

The European economy is highly dependent on imports of Ni, a raw material of high economic importance and agromining is an interesting technology for the recovery of this trace element from low-grade ores such as ultramafic soils. However, climatic conditions, soil properties and metal bioavailability can limit the production of plant biomass and metal accumulation, and consequently the effectiveness of agromining (van der Ent et al., 2015). In this context, one the objectives of the project LIFE-Agromine, funded by the EU, was the establishment of Ni agromining field scale demonstrations on distinct geographical areas and under different edapho-climatic conditions. The present study implemented agromining field trials in an ultramafic soil on the NW of Spain under a humid temperate climate for evaluating the performance of the native Ni hyperaccumulator *Odontarrhena serpyllifolia* and of the Mediterranean hyperaccumulators *Odontarrhena muralis* s.l. and *Bornmuellera emarginata*. The trials also aimed at comparing inorganic (NPK) and organic (cow manure) fertilisation and evaluated 3 cropping seasons. The highest amount of biomass (up to 8 t ha⁻¹) and Ni yield (40 kg ha⁻¹) was obtained with *O. muralis* cultivated with NPK fertilization. Inorganic fertilization was also the best option for the cultivation of *O. serpyllifolia*, although the harvested biomass and Ni yield were much lower than those of *O. muralis* s.l. Conversely, in the case of *B. emarginata* and in the 3 evaluated cropping seasons, the higher Ni yields (up to 11 kg ha⁻¹) were obtained in plots fertilized with cow manure. Interestingly, the harvested biomass of *B. emarginata* was similar in inorganic and organic fertilised plots. The cultivation of the Ni hyperaccumulators for 3 growing seasons did not significantly decrease the Ni availability. Plant ionome and other soil physic chemical parameters were also investigated and are presented.

KEYWORDS: *Ni hyperaccumulators, Ni yield, inorganic and organic fertilization, soil Ni availability*

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Phenanthrene impact on *Noccaea caerulescens* functioning and metal extraction efficiency - consequences for agromining on multi-contaminated soil

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ABSTRACT

Multi-contamination of soil by multiple organic and inorganic contaminants is considered an obstacle for the hyperaccumulator plants to develop and phytoextract metals. The aim of this study was to explore the impact of polycyclic aromatic hydrocarbons (PAHs) in combination with heavy metals on the phytoextraction efficiency and antioxidative response of the Ganges and Chavignée populations of the metal hyperaccumulator *Noccaea caerulescens*. Plants were cultivated for a duration of 17 days in a co-contaminated soil containing some heavy metals at moderate concentrations and spiked with phenanthrene (PHE), a model PAH, at a concentration of 200 mg PHE kg⁻¹ soil. The presence of PHE in the soil proved to be toxic for *N. caerulescens*. When exposed to PHE, plants exhibited typical stress response with a reduction in general growth parameters, along with the upregulation of antioxidant compounds and enzymes combined with limitations in nutrient uptake and heavy metals extraction. Variations were observed in the magnitude of enzymatic activities and the amount of extracted metals between the two studied populations. Nevertheless, to some extent, growth, metals extraction, and antioxidant defense responses differed slightly between the studied populations. Chavignée plants appeared slightly more tolerant to stress. This population is known to extract more Zinc (Zn) and Nickel (Ni) but less Cadmium (Cd) than the Ganges [1], suggesting that the difference in defense and extraction capacities might ensue different tolerance. This distinction may be related to the adaptations acquired by each population depending on the soil type it originated from.

KEYWORDS: *hyperaccumulator, PAH, abiotic stress, phytoextraction, multi-contaminated sites, antioxidants*

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Study of Different Shading Levels on the Growth of *Bornmuellera emarginata*: a Nickel Hyperaccumulator for Use in Agromining

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ABSTRACT

The Greek endemic *Bornmuellera emarginata*, known for its Ni hyperaccumulation capacity, is a promising candidate for use in agromining. Field observations that populations of this species usually occupy shaded places under shrubs or trees led us to investigate the effect of different shading levels [i.e. 0% (control), 20%, 35%, 60%, and 90%] on morphological and physiological parameters of *B. emarginata*. The experiment was conducted under greenhouse conditions and lasted 210 days after sowing (DAS). Seeds were sown directly into 2 L pots filled with serpentine soil collected from the field. Three plant samplings (81, 140 and 210 DAS) were conducted during the experiment for the determination of fresh and dry plant weight, plant height, number of the lateral shoots, total number of leaves per plant and proline concentrations. Chlorophyll concentrations in leaves were measured in nine samplings (64, 70, 81, 98, 104, 115, 140 and 210 DAS). As resulted, in the early stages plant growth was better under 20% and 35% shading, and the differences between the different shading levels being decreased by the end of the experiment. Plant height was increased while the number of the lateral shoots and the total number of leaves per plant were decreased as shading increased. At the end of the experiment (210 DAS), the lower proline concentration ($0.36 \mu\text{mol g}^{-1}$) was observed at 35% shading and the higher ($1.62 \mu\text{mol g}^{-1}$) at the control (0%). Leaf chlorophyll concentrations were increased as plants grew. The highest concentrations were observed at low shading (0%, 20% and 35%). At the end of the experiment (210 DAS), the highest values were 98.2 and $96.9 \mu\text{g cm}^{-2}$ at 35% and 60% shading, respectively while the lowest were 61.4 and $82.4 \mu\text{g cm}^{-2}$ in the control (0%) and at 20% shading, respectively. In the final sampling, nickel concentration of the aboveground part of the plants was determined.

KEYWORDS: *hyperaccumulator, shading, proline, chlorophyll*

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Effects of Irrigation with Treated Wastewater on the Nutrient Contents of *Salvia officinalis*

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ABSTRACT

This article aims to provide information on the evaluation of nutritional skill in the *Salvia officinalis* L. plant after its irrigation with treated wastewater. Field experiment was chosen as the main method of collecting information, and was conducted to determine the effect of water quality on the plant (treated wastewater, untreated wastewater and well water), as well as the interactions of these waters on the growth and water use efficiency in the first year of vegetation growing of *S. officinalis* L. This plant was chosen as a plant that is cultivated in dry areas and that is not part of the food chain. The effects of irrigation with treated wastewater (TWW) were compared with those of well waters (WW) at the level of nutrient elements. Referring to DM (dry matter), the percentage of total nitrogen in the plant leaf irrigation with treated wastewater was classified at the highest level, up to 2.6%. Whereas, in the plants irrigated with well water, the nitrogen content level varies from 1.5 to 3.5%. Likewise, Phosphorus levels varied from 0.1 to 0.3%, while for the nitrates form a significant increase was reported in stems of plants irrigated with treated water at the level of 0.4%. This was attributed to the mobility of this form. The concentration levels of ammoniacal form found in this plant ranged from 61.46 mg/kg to 184.1 mg/kg, which being an immobile and positive charged form, is kept form the absorbing earth complex. The results of the experiment confirmed the efficiency of recycled wastewater use, while not negatively affecting the quality of the product of *Salvia officinalis* L.

Keywords: *Treated wastewater, well water, Salvia officinalis* L., *macronutrients (nitrogen, phosphorus) etc.*

Designing Low Cost Cropping Systems for Nickel hyperaccumulators in the Balkans

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After the LIFE Agromine project (2016-2021), the techno-economic assessment emphasized on the fact that, to be viable, operations had to be mechanised or carried out in countries where labour cost is low enough to allow viable implementation of the crops [1]. In order to implement commercial agromining in EU countries, it is therefore crucial to mechanise all the steps of the crops and therefore minimise their costs. New cropping systems are proposed here which are adapted to all regions in the Balkans for three main species, which proved to be successful in the LIFE Agromine project: *Odontarrhena chalcidica* (syn= *Alyssum murale*), *Bornmuellera emarginata* (syn= *Leptoplax emarginata*) and *Bornmuellera tymphaea*.

Odontarrhena chalcidica are native to all ultramafic areas in the Balkans and hence do not require the production of seeds beforehand. Seeds can be spontaneously produced on every agricultural field that is regularly ploughed. It is ideal to select fields on which substantial covers of *Odontarrhena chalcidica* have seeded in the summer before ploughing. Plots are then ploughed in winter, seedstock in the soil will create a spontaneous cover in the spring that needs to be partially weeded mechanically (to create rows, ideally every 50 cm). Covers can be fertilised (50 kg N) at the beginning of October to enhance further growth and branching of plants. In early spring, the plot is again fertilised (NPK 15–15–15 fertilizer at the rate of 100 kg per ha) and harvested at early flowering stage with a conventional hay mower, then baled, usually at the beginning of June.

For *Bornmuellera* species, it is crucial to rely on perennial plant covers, due to the high cost of transplanting 40,000 seedlings per ha. For instance, for *B. emarginata*, those phenotypes found in nature that are fully perennial need to be selected (breeding). *Bornmuellera tymphaea* is a fully perennial species. Both species can be easily propagated by softwood cuttings that are prepared in a nursery over winter and early spring. *Bornmuellera emarginata* can also be sown directly in the field with a seeder. Ideally, they should be transplanted in April to ensure sufficient growth of the plants before summer. *Bornmuellera emarginata* can be harvested twice a year to stimulate branching, once at the beginning of October and the second time in spring at early flowering (May). Both plant are fertilised in the same way as *O. chalcidica*. Harvest is carried out in the same way as for medicinal and aromatic plants (i.e. Oregano).

KEYWORDS: phytomining, agromining, nickel, hyperaccumulators, cropping systems

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Thank you for your participation!

