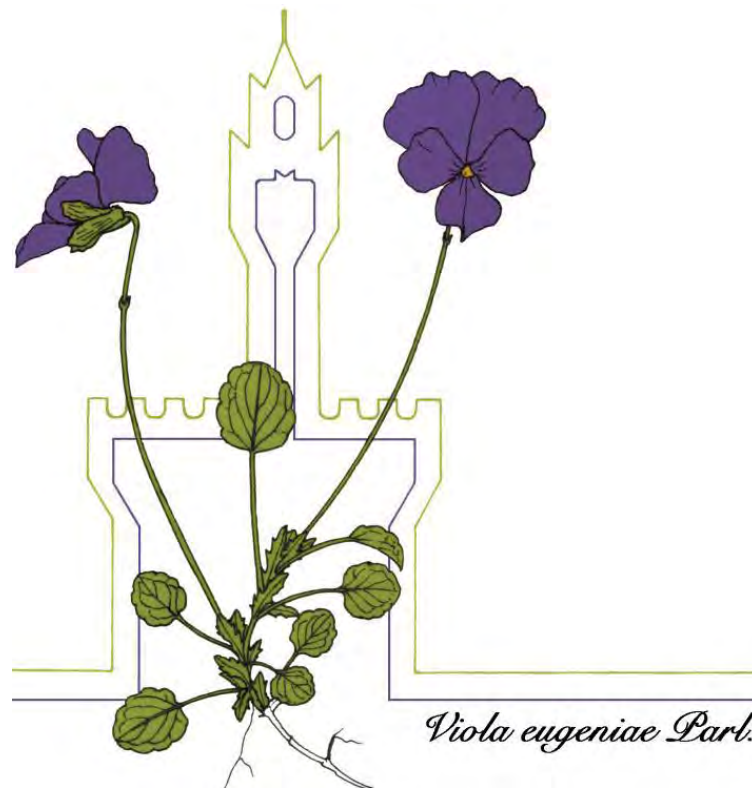


Società Botanica Italiana

120° Congresso



XI International Plant Science Conference (IPSC)

GoBot2025

Overcoming Barriers in Plant Science and Beyond

Gorizia, September 3-6, 2025

ABSTRACTS

KEYNOTE LECTURES, COMMUNICATIONS, POSTERS

Organized by



Patronized by:



Supported by:



Scientific Committee:

Antonella Canini (Università di Roma Tor Vergata), Barbara Baldan (Università di Padova), Anna Maria Mercuri (Università di Modena e Reggio Emilia), Gianni Sacchetti (Università di Ferrara), Giovanni Spampinato (Università di Reggio Calabria), Roberto Venanzoni (Università di Perugia), Giuseppe Venturella (Università di Palermo), Mauro Tretiach (Chair, Università di Trieste), Jože Bavcon (University of Lubiana), Francesco Boscutti (Università di Udine), Stefano Martellos (Università di Trieste), Giuseppina Alongi (Università di Catania), Gianni Bedini (Università di Pisa), Giulia Caneva (Università di Roma Tre), Daniela Ciccarelli (Università di Pisa), Annalena Cogoni (Università di Cagliari), Gianniantonio Domina (Università di Palermo), Gabriele Galasso (Comune di Milano), Marta Galloni (Università di Bologna), Salvatore Lenucci (Università del Salento), Guido Lingua (Università del Piemonte), Stefano Loppi (Università di Siena), Alessia Masi (Sapienza Università di Roma), Simone Orsenigo (Università di Pavia), Alessio Papini (Università di Firenze), Ferrucci Poli (Università di Bologna), Leonardo Rosati (Università della Basilicata), Francesco Tarantino (Lecce), Solveig Tosi (Università di Pavia).

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Web site: please visit the web pages at
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120° Congresso della Società Botanica Italiana onlus

XI INTERNATIONAL PLANT SCIENCE CONFERENCE (IPSC)

GoBot2025

Overcoming Barriers in Plant Science and Beyond

Gorizia, September 3-6, 2025

Programme

Tuesday, September 2nd

For the conveners arriving in Gorizia in the evening:

19:00-21:00 **Welcome Cocktail - location TBD**

Wednesday, September 3rd

Venue: Auditorium della Cultura Friulana, Via Roma 23

08:30 - 09:30 Registration

09:30 - 10:30 **Symposium in memory of Livio Poldini - Modern biogeography. Studying and classifying plant ranges and communities, I part**

Chair persons: **Roberto Venanzoni e Mitja Caligarič**

Invited lecture: **Mitja Kaligarič** (25 minutes + 5 minutes for discussion) - Poldini's Inspirations for Research in the Slovenian Karst: Linking Plant Communities, Grassland Ecology, Vegetation History, and Human Impact

Communications (10 minutes + 2 minutes for discussion):

- **Giacomo Baldesi**, Fabio Palumbo, Daniela Calzoni, Giulia Ginelli, Simone Orsenigo - Phylogeography and phylogenomics of the *Hieracium tomentosum* group
- **Andraž Čarni**, Aljaž Jakob, Mateja Breg Valjavec - Dolines on Karst Plateau as an object of vegetation studies
- Federico Selvi, Roberto Canullo, **Elisa Carrari**, Giandiego Campetella, Stefano Chelli, Andrea Coppi, Emmanuele Farris, Alfredo Maccioni, Francesco Mascia, Nicola Postiglione, Leonardo Rosati, Camilla Wellstein, Cristina Gasperini - Leaf trait variation across Italian forest endemics: drivers and divergence from widespread congeners

- **Marco Cervellini**, Giandiego Campetella, Stefano Chelli, Luciano Ludovico Maria De Benedictis, Giorgio Brunialti, Antonella Canini, Alessandro Chiarucci, Piermaria Corona, Rosa M. Di Biase, Domenico Di Martino, Lorenzo Fattorini, Arianna Ferrara, Sara Franceschi, Luisa Frati, Giovanni Jacopetti, Agnese Marcelli, Marzia Marcheselli, Walter Mattioli, Giancarlo Papitto, Caterina Pisani, Federico Selvi, Roberto Canullo - The Italian National Forest Inventory (IFNI2025): a new opportunity for plant diversity monitoring
- **Luca D'Amato**, Arturo Gallia, Valentina Savo - Plant biodiversity patterns and conservation challenges in Mediterranean minor islands: a comprehensive review
- **Hicham El Zein**, Mauro Fois, Benedetta Gori, Gianluigi Bacchetta - Endemism patterns of the vascular flora of Lebanon and ex situ conservation aspects
- **Jacopo Franzoni**, Giuseppe Antonelli, Marco D'Antraccoli, Lorenzo Peruzzi - Peripheral population or understudied distribution? The tale of *Crepis suffreniana* (Asteraceae) in Italy

10:30 - 11:00 Coffee break

11:00 - 12:00 **Symposium in memory of Livio Poldini - Modern biogeography. Studying and classifying plant ranges and communities, II part**

Chair persons: **Mitja Caligarić, Roberto Venanzoni**

Communications (10 minutes + 2 minutes for discussion):

- **Sara Natale**, Marco Canella, Garima Singh, Mariano Battistuzzi, Silvia Moschin, Anna Pasinato, Valentina Boscarol, Barbara Baldan, Francesco Dal Grande, Nicoletta La Rocca, Alessandro Alboresi - Leaf spectral properties and leaf traits of four alpine species of butterworts (*Pinguicula* L.)
- **Simone Orsenigo**, Andrea Bianchi, Silvia Ceppi, Giacomo Baldesi - Ngezi-Vumawimbi Forest Reserve, Pemba Island, Zanzibar (Tanzania): a unique biogeographical hotspot
- **Veronica Ranno**, Salvatore Cambria, Pietro Minissale, Gianpietro Giusso del Galdo - Diachronic analysis of Etna high-mountain plant communities
- **Giulia Tomasi**, Giacomo Trotta, Michael Kleih, Alessio Bertolli, Enzo Bona, Roberto Dellavedova, Alberto Selvaggi, Francesco Boscutti - Floristic and Ecological Characterization of Endemic Plants in Northern Italy: spatial patterns and environmental drivers

12:00 - 13:00 **Opening ceremony and institutional greetings, with a short lecture entitled: Gorizia, borderland.**

13:00 - 14:30 **Lunch break**

Location: Wienere Haus Gorizia (2-3 minutes walk from the Congress venue)

Parallel sessions

Venue: Auditorium della Cultura Friulana, Via Roma 23

14.30 - 16:00: Forest communities: from management to conservation

Chair persons: Roberto Canullo, Giovanni Spampinato

Communications (10 minutes + 2 minutes for discussion):

- **Iduna Arduini**, Matteo Diana, Riccardo Lenci - Ellenberg Indicator Values demonstrate community shift and habitat loss in the forest of San Rossore (Nature 2000 Site 'Selva Pisana')
- **Sara Barberini**, Alberto Bernacchi, Gianni Della Rocca, Roberto Danti, Lorenzo Arcidiaco, Lorenzo Cotrozzi, Giovanni Emiliani - The mycobiome of declining holm-oak populations of the Maremma Regional Park (Italy): Insight into fungal community dynamics and potential drivers of tree decline
- **Marta Elisabetta Eleonora Temporiti**, Silvia Traversari, Lidia Nicola, Alessio Giovannelli, Sara Barberini, Giovanni Trentanovi, Solveig Tosi, Maria Laura Traversi, Giovanni Emiliani - Influence of the management of alpine Norway spruce forests on plant and soil mycobiomes
- **Luciano Ludovico Maria De Benedictis**, Stefano Chelli, Zhengxue Zhu, Marco Cervellini, Roberto Canullo, Francesco Chianucci, Nicola Puletti, James Lee Tsakalos, Sándor Bartha, Giandiego Campetella - Multiscale assessment of understory functional diversity components and their relationships with forest structure and canopy cover
- Raffaello Resemini, Andrea Arcidiacono, Andrea De Toni, Sandra Citterio, Cinzia Panigada, Micol Rossini, Luigi Vignali, **Rodolfo Gentili** - Assessment of landscape, structural and biodiversity metrics of afforestation actions over the last 50 years in the Milan Metropolitan area
- **Lorenzo Orzan**, Antonio Tomao, Luca Cadez, Valentino Casolo, Paolo Cingano, Claire Duvaltier, Roberta Pascolo, Giacomo Trotta, Giorgio Alberti - Linking forest structure and plant diversity across secondary forest succession
- **Ilaria Santi**, Elisa Carrari, Pieter Sanczuk, Pieter Vangansbeke, Karen De Pauw, Thomas Vanneste, Kurt Bollmann, Jörg Brunet, Kim Calders, Sara AO Cousins, Martin Diekmann, Cristina Gasperini, Bente J Graae, Paige Heavyside, Sigrid Lindmo, Per-Ola Hedwall, Giovanni Iacopetti, Jonathan Lenoir, Anna Orczewska, Quentin Ponette, Jan Plue, Markus Bernhardt-Römermann, Federico Selvi, Fabien Spicher, Thomas Vanneste, Hans Verbeeck, Liping Wei, Florian Zellweger, Pieter De Frenne - Shifts in understory diversity and thermophilization under climate change are modulated by canopy buffering: evidence from a 5-years re-survey study across European forests

16.00-16.30 Coffee break

16.30-18.00: **Mutualism and competition in a changing world**

Chair persons: **Anna Maria Mercuri, Lucia Muggia**

Invited lecture: **William B. Sanders** (25 minutes + 5 minutes for discussion) - Lichen Symbioses: Partnerships of Contingency

Communications (10 minutes + 2 minutes for discussion):

- **Enrico Boccato**, Davide Porrelli, Claudio Gennaro Ametrano, Fabio Candotto Carniel, Mauro Tretiach - When *Trebouxia* swims: flagellate cells and hidden sex in the life cycle of a lichen photobiont
- **Chiara Buonanno**, Giovanna Aronne - Detailed floral traits widen the discussion on pollination strategy of *Papaver rhoeas* L. (Papaveraceae)
- **Alfredo Maccioni**, Emmanuele Farris - Plant-plant and plant-animal interactions in the recruitment of *Juniperus turbinata* Guss. in Sardinian rocky coastal habitats
- **Samuele Pignedoli**, Daniele Calabrese, Elena Tafi, Matteo Dal Cin, Gianni Henson, Massimo Nepi - Yeast diversity in *Onosma* spp. floral nectar and pollinators
- **Daniela Zuzolo**, Gina Gizzi, Maria Tartaglia, Antonello Prigioniero, Maria Antonietta Ranauda, Maria Maisto, Monica Labella-Ortega, Emanuele Fosso, Amy Ellison, Simon Creer, Carmine Guarino - Landscape complexity shapes plants-arthropods interactions in agroecosystems: an eDNA approach

Venue: Centro Polifunzionale dell'Università di Udine, Via Santa Chiara 1

14:30 - 16:00 **Botanic collections in the 21st century**

Chair persons: **Gianni Bedini, Stefano Martellos**

Invited lecture: **Heimo Rainer** (25 minutes + 5 minutes for discussion) - Prospects for and requirements of modern botanical collections in a global context.

Communications (10 minutes + 2 minutes for discussion):

- **Lorenzo Braga**, Giacomo Vinci, Eleonora Rattighieri, Assunta Florenzano, Alessandro Fontana, Anna Maria Mercuri - Palaeobiodiversity of wetlands among buried protected biotopes and glacial relicts: preliminary palynological findings and time-based indicator species from the Teor-Campomolle palaeochannel sedge peatland (NE Italy)
- **Mónica Labella-Ortega**, Guido Cipriani, Rosario Schicchi, Giuseppe Bazan, Hengchi Chen, Maria Antonietta Ranauda, Maria Maisto, Daniela Zuzolo, Maria Tartaglia, Antonello Prigioniero, Carmine Guarino - The evolutionary history of the endemic palm *Chamaerops humilis* L. in the Mediterranean
- **Elia Lipreri**, Enzo Bona, Mario Ferrari, Alessio Mereu, Stefano Armiraglio - The Herbarium of Brescia: Digitization, Virtual Access and Future Perspectives
- **Weiwei Liu**, Jacopo Zacchigna, Felice Andrea Pellegrino, Adriano Peron, Francesco Roma-Marzio, Lorenzo Peruzzi, Stefano Martellos - Application of computer vision to the

automated extraction of metadata from natural history specimen labels: A case study on herbarium specimens

- **Yasaman Ranjbaran**, Marco Canella, Sara Natale, Anna Pasinato, Garima Singh, Sven Winter, Francesco Dal Grande - Tracking genomic shifts in alpine flora: a museomics approach to climate change assessment

16.00-16.30 **Coffee break**

16:30 - 17:00 **Botanic collections in the 21st century**

Chair persons: **Gianni Bedini, Stefano Martellos**

Communications (10 minutes + 2 minutes for discussion):

- Jože Bavcon, **Blanka Ravnjak** - Green infrastructure as an option for *ex-situ* plant conservation
- **Fabio Rindi**, Anna Maria Mannino, Simona Armeli Minicante - Italian macroalgal herbaria in the 21st century: present uses and new roles in modern phycological research
- **Linda Seggi**, Lorenzo Cecchi - Historic herbaria as cultural heritage: pre-Linnaean collections in Italian institutions

17:00 - 18:00 **Overcoming discipline barriers: Art and plant sciences**

Chair persons: **Franco Palla, Lorenzo Peruzzi**

Communications (10 minutes + 2 minutes for discussion):

- **Giulia Caneva**, Alessandro Lazzara, Alma Kumbaric - The hidden botanical language in the ancient Roman architecture: the example of the Adrian's Arch at Jerash (Jordan)
- **Sofia Ceseri**, Ornella Salvadori, Francesco Princivale, Barbara Salvadori, Mauro Tretiach - Occurrence of the lichen *Tephromela atra* on carbonate-rich rocks: what's so strange?
- **Sergio E. Favero Longo**, Marta Cicardi, Laura Guidorzi, Enrica Matteucci, Mariagrazia Morando, Daniele Castelli, Diego Elia - Lichen recolonization on stone heritage surfaces following cleaning interventions: Should preventive strategies already consider the ongoing climate change?
- **Roberta Ranaldi**, Lorenza Rugnini, Francesco Gabriele, Gerardo Pepe, Nicoletta Spreti, Patrick Di Martino, Giada Migliore, Flavia Tasso, Francesco Scuderi, Roberto Braglia, Pujia Angelica, Antonella Canini - The Colosseum in green – Can essential oils promote sustainable conservation?
- **Silvia Del Vecchio**, Gabriele Gheza, Juri Nascimbene, Fabrizio Buldrini - Reconstructing coastal dune flora from 400 years ago: insights from the Aldrovandi's herbarium.

Venue: **TBD**

18.30-20:00 **AperiWorkshop!**

Format: Ten minutes presentation per project, and free discussion.

The following project proposals will be presented:

- **Mauro Fois** - Towards a better knowledge about the distribution of the old and notable trees of Italy: a citizen science approach
- **Jacopo Franzoni**, Antonio Giacò, Lorenzo Peruzzi - Estimating the biological support of species hypotheses in the Italian native flora
- **Linda Seggi**, Lorenzo Cecchi - Towards a national framework for historical scientific collections: mapping standardizing and digitizing Italy's Pre-Linnaean herbaria

Venue: **Auditorium della Cultura Friulana, Via Roma 23**

20:30 - 22:30 **Public event (in Italian) - Piante, confine tra terra e cielo: Impatto dei cambiamenti climatici ai limiti della vita.**

Partecipano:

Antonella Canini (Presidente della Società Botanica Italiana, Università di Roma "Tor Vergata")

Alessandro Chiarucci (Università di Bologna "Alma Mater")

Piero Genovesi (ISPRA)

Luca Mercalli (Società Meteorologica Italiana)

Alessandro Petraglia (Università di Parma)

Modera: **Marco Virgilio** (Giornalista)

Thursday, September 4th

Venue: **Centro Polifunzionale dell'Università di Udine, Via Santa Chiara 1**

09:00 - 10:30 **Poster session 1-3** (3 parallel sessions) - 2 minutes presentation plus 1 minute discussion

Venue: Auditorium della Cultura Friulana, Via Roma 23

10:30 - 11:00 Coffee break

11:00 - 13:00 Plants overcoming barriers: native and alien plants facing global changes

Chair persons: Sandra Citterio, Adriano Stinca

Invited lecture: Franz Essl (25 minutes + 5 minutes for discussion) - The global re-shuffling of species

Communications (10 minutes + 2 minutes for discussion):

- **Sarah Caronni**, Nicoletta Ancona, Lara Assunta Quaglini, Chiara Montagnani, Claudia Russo, Marco Tamburini, Sandra Citterio - Temperature-driven dynamics of *Caulerpa taxifolia* (Vahl) C.Agardh epiphytic bacterial community: implications for the alien alga diffusion in a changing climate
- **Paolo Cingano**, Elisa Pellegrini, Stefano Tomat, Edoardo Asquini, Giacomo Boscarol, Giacomo Trotta, Marco Vuerich, Francesco Boscutti - Functional traits and plant-soil feedback mechanisms contribute to the understanding of spatial pattern formation of *Cymodocea nodosa* meadows
- **Mauro Fois**, Alba Cuena-Lombraña, Jennifer Nagel Boyd, Lina Podda, Gianluigi Bacchetta - Investigating habitat and functional traits of nativeness, rarity, and invasiveness in Sardinia's aquatic plants
- **Fabio Mologni**, Peter J. Bellingham, Ewen K. Cameron, Anthony E. Wright - Time since first naturalization is key to explaining non-native plant invasions on islands
- **Chiara Montagnani**, Nicole Sebesta, Emanuele Vegini, Giulia Daniele, Elena Barni, Laura Celesti-Grapow, Francesca Emili, Federica Larcher, Rodolfo Gentili, Maria Laura Carranza, Angela Stanisci, Lucia Antonietta Santoianni, Marco Varricchione, Sandra Citterio - Urban jungle: mapping the spread of invasive alien plants across Italian cities
- **Carmelo Maria Musarella**, Antonio Morabito, Serafino Cannavò, Valentina Lucia Astrid Laface, Ana Cano Ortiz, Sara Del Rio, Ricardo Quinto Canas, Giovanni Spampinato - Counteracting the invasion of *Cenchrus setaceus* (Forssk.) Morrone for the recovery of native vegetation: an eradication project from Calabria (southern Italy)
- **Marco Vuerich**, Edoardo Asquini, Giacomo Boscarol, Enrico Braidot, Michele Carbognani, Giorgio Chiari, Paolo Cingano, Arianna Del Pino, T'ai GW Forte, Filippo Grillo, Alessandro Petraglia, Giacomo Trotta, Marco Zancani, Francesco Boscutti - The role of duration and timing of induced heat waves in shaping plant community of alpine grasslands: an upscaling approach

13:00 - 14:30 Lunch break

Location: Wienere Haus Gorizia (2-3 minutes walk from the Congress venue)

Venue: Auditorium della Cultura Friulana, Via Roma 23

14:30 - 16:30 Cell biology and specialized metabolites

Chair persons: Barbara Baldan, Gianni Sacchetti

Communications (10 minutes + 2 minutes for discussion):

- **Gaia Cusumano**, Giancarlo Angeles Flores, Mehmet Veysi Cetiz, Umran Kurt, Gunes Ak, Enver Saka, Shaza H. Aly, Omayma A. Eldahshan, Abdel Nasser Singab, Gokhan Zengin, Ismail Senkardes, Maria J. Rodrigues, Luisa Custodio, Carla Emiliani, Paola Angelini - From Folk Medicine to Functional Applications: Phenolic Composition and Bioactivity of *Stellaria media*
- **Paola Frazzetto**, Elena Bombaci, George W Bassel, Antonia Cristaudo, Giuseppe Diego Puglia - Molecular mechanisms underlying seed germination ecophysiology of *Cardamine occulta* Hornem. (Brassicaceae) in relation to ROS production
- **Giulia Giorgi**, Aylin Sahin, Adriano Patriarca, Emanuele Zannini, Elisa Brasili - Nutritional properties, chemical characterization and techno-functional analysis of *Cyperus rotundus* L. tubers
- **Aurora Kupe**, Maria Eleonora Foletti, Gianni Sacchetti, Massimo Tacchini - From *Cannabis sativa* L. (Cannabaceae) by-products to bioactive agents: extraction and biological potential
- **Eileen Mac Sweeney**, Vlad Sebastian Popescu, Gianluca Angius, Ylenia Pieracci, Guido Flamini, Maurizio Memo, Luisa Pistelli, Andrea Mastinu - Antioxidant properties of three *Lavandula* genus essential oils compared to a commercial essential oil
- **Katja Malovrh**, Mitja Križman, Blanka Ravnjak, Jože Bavcon - Analysis of sugars, amino acids and phenolic compounds in nectar of selected melliferous native species in Slovenia
- **Elisabetta Offer**, Silvia Moschin, Emanuela Talarico, Leonardo Bruno, Barbara Baldan - Reproductive mechanisms in *Ginkgo biloba*: male cone development, pollen formation, and the intricate crosstalk between male and female gametophytes
- **Alice Peduzzi**, Diego Piacentini, Elisabetta Speranza, Tiziano Napoleoni, Federica Della Rovere, Simone D'Angeli, Nitin Pawar, Peter Nick, Maria Maddalena Altamura, Giuseppina Falasca - Brassinosteroids seed-priming to enhance *Sorghum bicolor* salt tolerance
- **Francesca Rapparini**, Gabriella Vinci, Alessio Tei, Gergana Mihailova, Katya Georgieva - Resurrection Plants and their Volatilome: the unresolved mystery of BVOC emissions under extreme desiccation
- **Sara Vitalini**, Orlando Innaurato, Shadab Famarzi, Alessandro Palmioli, Stefania Garzoli, Valentina Vaglia, Cristina Airoldi, Marcello Iriti - Phytotoxicity of *Zingiber officinale* Roscoe aerial biomass: Bioactive potential for sustainable weed suppression

16:30 - 17:00 **Coffee break**

17:00 - 18:30 **General Meeting of the Italian Botanical Society (members only)**

18:30 - 20:00 **Guided tour of the city center, with an introduction to the historic and societal relevance of Gorizia in a bordering area**

20:30 **Congress social dinner**

Location: Wiener Haus Gorizia (2-3 minutes walk from the Congress venue)

Friday, September 5th

Venue: Centro Polifunzionale dell'Università di Udine, Via Santa Chiara 1

09:00 - 10:30 **Poster session 4-6** (3 parallel sessions) - 2 minutes presentation plus 1 minute discussion

Venue: Auditorium della Cultura Friulana, Via Roma 23

10:30 - 11:00 **Coffee break**

11:00 - 13:00 **Photoautotrophs under shifting water regimens; what's new?**

Chair persons: Giuseppina Alongi, Lorenzo Ferroni

Invited lecture: Nate McDowell (25 minutes + 5 minutes for discussion) - The future of critical transitions at leaf to community scales

Communications (10 minutes + 2 minutes for discussion):

- **Mario Di Stefano**, Cristina Blandino, Eduardo Fernández-Pascual, Giuseppe Diego Puglia, Antonia Cristaudo - Differentiation of seed germination traits along a small-scale hydroperiod zonation in Mediterranean temporary ponds
- **Riccardo Fedeli**, Stefano Loppi - Wood distillate as a biostimulant for mitigating drought stress in basil: effects on growth, physiology, and nutrient balance

- **Andrea Morettini**, Sanchez Sergio, Sergi Garcia Barreda, Mara Rondolini, Leonardo Baciarelli Falini, Andreea Daniela Dam, Diletta Bisonni, Domizia Donnini - Comparison of morphological and molecular analysis method for truffle plants used in Spain, France and Italy
- Alberto Danieli, Peter Beatrice, Tsvetana Masante, **Chiara Pezzuto**, Angela Balzano, Gabriella Stefania Scippa, Antonio Montagnoli - Effect of Heat, Drought, and their interplay on Stem and Root Cambium cell activity in *Populus nigra* L.
- **Alessio Tei**, Lucía Martín-Cacheda, Xoaquín Moreira, Sergio Rasmann, Gregory Röder, Irene Martínez-Virveda, Carla Vázquez-González - Precipitation-driven variability in VOC and SVOC emissions in the carnivorous plant *Drosera rotundifolia* L. (Droseraceae)

13:00 - 14:30 **Lunch break**

Location: Wienere Haus Gorizia (2-3 minutes walk from the Congress venue)

Venue: Auditorium della Cultura Friulana, Via Roma 23

14:30 - 16:30 **Overcoming barriers and borders in plant monitoring, conservation, and ecological restoration**

Chair persons: **Gianluigi Bacchetta, Simone Orsenigo**

Communications (10 minutes + 2 minutes for discussion):

- **Sonia Ravera**, Renato Benesperi, Juri Nascimbene, Marta Agostini, Luca Di Nuzzo, Elisabetta Bianchi, Patrizia Campisi, Silvia Del Vecchio, Giovanna Pezzi, Monica Ruffini Castiglione, Luigi Sanità di Toppi, Luca Paoli - Integrating Seasonal Translocation Strategies for the Conservation of *Lobaria pulmonaria* in Mediterranean Woodlands: Insights from the BioConLobaria Project
- **Valentino Casolo**, Paloma Perez-Bello, Fabio Marroni, Emanuele De Paoli, Giuseppe Fabrini, Lara Dixon, Roberta M. Ceriani, Luciano di Martino, Blanka Ravnjak, Joseph Buhagiar, Gianluigi Bacchetta, Antonia Cristaudo, Gabriele Casazza, Cristina Salmeri, Mariacristina Villani, Sara Magrini, Costantino Bonomi - ddRAD sequencing: a valuable method for assessing genetic diversity in plant conservation projects
- **Emma Cocco**, Ivan Blečić, Eleonora Buoio, Annalena Cogoni, Paolo Colleto, Antonio De Agostini, Ferdinando Fornara, Giulia Giliberto, Andrea Manunza, Oriana Mosca, Emanuel Muroni, Giulia Nuscis, Pierluigi Cortis - TERSICORE Project: a multidimensional approach to urban greening, environmental monitoring, and community engagement for Urban Regeneration
- **Diana María Cruz-Tejada**, Efisio Mattana, Angelino Carta - Modelling seed germination data to meet conservation needs in Mediterranean habitats
- **Azzurra Di Bonaventura**, Paolo Cingano, Francesco Boscutti, Michele Parioli, Behixhe Ajdini, Stefano Accoroni, Anna Annibaldi, Edi Gljušćić, Andrea Bilajac, Ljiljana Iveša, Mirta Smodlaka Tankovic, Luca Gemolotto, Francesco Trevisan, Ivan Scagnetto, Monica

Ortega Mosquera, Sabina Susmel, Fabio Rindi - The BRIGANTINE project: a new sustainable approach for monitor the health status of seaweeds and seagrasses in the North Adriatic Sea

- **Giovanni Emiliani**, Sara Barberini, Gianni Della Rocca, Rosario Schicchi, Roberto Danti - The mycobiome of the critically endangered *Abies nebrodensis*: environmental conditions are the drivers of fungal diversity
- **Jana Gerstmayr**, Gerald M Schneeweiss, Jalil Noroozi - Hotspots of Alpine Vascular Plants in the Andes and Their Conservation Status
- **Stefano Martellos**, Linda Seggi, Raffaella Trabucco, Annalisa Falace, Alessandra Metalli, Riccardo Fedeli, Stefano Loppi - Biomonitoring of potentially toxic elements in the Venice Lagoon (Italy): a 100-year comparison using museum specimens
- **Staš Miljuš**, Polona Božič, Aleksander Kozina, Blaž Blažič, Katarina Denac - Decline of High Nature Value Grassland Habitats Within Natura 2000 Sites in Slovenia
- Elisabetta Sgarbi, **Aya Mortada**, Gioele Toselli, Niccolò Patelli, Lara Maistrello - Monitoring of wild orchids in urban and natural ecosystems and their potential pollinators
- **Maria Tartaglia**, Monica Labella Ortega, Antonello Prigioniero, Daniela Zuzolo, Carmine Guarino - Advanced Monitoring of Phytoremediation Effectiveness in Multi-Contaminated Sites through Metaproteomic approach

16:30-17.00 **Introduction to the excursion**

17:00 **Congress Closure**

Venue: **TBD (co-organized with the Municipality of Gorizia)**

18:00 - 19:30 - **AperiScience**. Evento pubblico di comunicazione sulla biodiversità urbana e peri-urbana (in Italiano).

Saturday, September 6th

09:00 - 16:00 **Field trip in the Karst**

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Poldini's Inspirations for Research in the Slovenian Karst: Linking Plant Communities, Grassland Ecology, Vegetation History, and Human Impact I

SYMPOSIUM 3

MUTUALISM AND COMPETITION IN A CHANGING WORLD

William B. Sanders
Lichen Symbioses: Partnerships of Contingency II

SYMPOSIUM 4

BOTANIC COLLECTIONS IN THE 21ST CENTURY

Heimo Rainer
Prospects for and requirements of modern botanical collections in a global context III

SYMPOSIUM 6

PLANTS OVERCOMING BARRIERS - NATIVE AND ALIEN PLANTS FACING GLOBAL CHANGES

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IN MEMORY OF LIVIO POLDINI - MODERN BIOGEOGRAPHY

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KEYNOTE LECTURES

Poldini's Inspirations for Research in the Slovenian Karst: Linking Plant Communities, Grassland Ecology, Vegetation History, and Human Impact

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Poldini's research in vegetation science and biogeography in the 1980s and 1990s inspired many Slovenian botanists and ecologists to conduct studies in the Slovenian Karst region and, more broadly, across the Alps, Istria, and the Dinarids. They adopted his methodologies, approaches and vegetation classification systems, which provided a robust foundation for the expansion of their research and initiated new directions in vegetation science. Research, conducted at the University of Trieste, combined with personal connections, mentorship, and joint field excursions with Slovenian botanists, also proved invaluable for our group at the University of Maribor.

Starting with new classifications of syntaxa and crosswalks to habitat typologies, this progression fostered diverse research directions in the Slovenian Karst. The floristic approach in phytosociology was supplemented by a functional approach, where in-situ measurements of plant traits of several hundred grassland species in the region were carried out. This enabled functional comparisons across biogeographical, geological and altitudinal gradients, as well as between different land management types (meadows vs. pastures) and significantly enriched the floristic approach. Consequently, it became possible to define plant guilds and CSR strategies, contributing to a better understanding of species coexistence, dominance and complementarity in resource acquisition within a given community. The functional approach was further advanced by phylogenetic approach and eco-physiological studies. The role of transhumance in the Karst was examined by studying propagules, transported in sheep fur; over 100 grassland species exhibited a few distinct dispersal strategies, facilitated by sheep movement.

The vegetation history of the Karst region was reconstructed using 250-year-old cartographic sources, extending to detailed field mapping and agricultural land-use records from recent decades. One study demonstrated that grasslands, which originally covered 82% of the area, have shrunk to 22% (including unmanaged grasslands) or to just 17% (only managed grasslands).

Using two historical temporal windows, spatial modelling algorithms were initially tested and subsequently refined. This enabled projection of vegetation changes up to the end of the 21st century. Thus, spatial variability in vegetation change patterns across the region was also obtained. By incorporating evapotranspiration data into vegetation cover maps, forest progression could be visualized in three dimensional scale, allowing detection of successional stages (scrub, young forest, mature forest) and the impact of sleet or fire events.

Following Slovenia's accession to the EU, the introduction of common legal framework has given new impetus to the protection of species and habitats. Species richness and below- and above-ground biomass were assessed under four sheep grazing regimes. Results showed that light grazing promotes the highest species richness and favours investment in below-ground biomass, which benefits habitat specialists. However, the age structure of farm owners, fragmentation of agricultural land, and abandonment of livestock farming are accelerating the decline of species-rich Natura 2000 grasslands. A methodological framework has been developed to support the monitoring and planning of agricultural activities and nature conservation measures at the regional level.

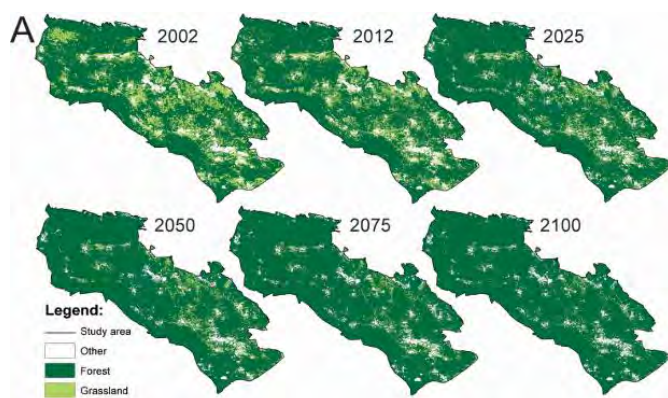


Fig. 1. Prediction of vegetation cover by 2100.



Fig. 2. Seeds, excavated from sheep wool.

Lichen Symbioses: Partnerships of Contingency

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Lichens have been within the purview of botany since Linnaeus conceived of a “vegetable kingdom,” a concept that is still surprisingly influential despite its biological heterogeneity and phylogenetic nonsensicality. Schwendener’s revelation that lichens are not in fact unitary organisms but rather composites of fungus and alga contributed to the subsequent introduction of the symbiosis concept. In a natural world seen increasingly from the perspective of Darwinian competition, symbiosis was viewed as somewhat of an ecological curiosity until its fundamental, recurring role in eukaryote macroevolution became widely appreciated in the late 20th century. Multiple and sequential endosymbiotic events repeatedly turned heterotrophic lineages into algae and plants, with symbionts gradually consolidated as organelles within cells of what are now recognized as unitary organisms. Lichens are also examples of heterotrophic lineages transformed by incorporation of photosynthetic symbionts, but pose particular conceptual challenges because the symbiosis does not approach the point of full and irrevocable union. Despite the remarkable integration of the symbionts structurally and functionally, they remain very much independent organisms at the cellular, genetic and phylogenetic levels. Collaboration of the symbionts is only achieved under conditions suboptimal for their separate development. While vegetative propagules containing both symbionts are commonly produced and dispersed, they likely also give rise to free-living individuals where microsite conditions are unsuitable for collaborative development. Reentry into lichen symbiosis involves establishment of a contact zone sealed with hydrophobic materials to allow substances to be exchanged between symbiont cells. The morphology of the contact zone is highly variable among lichen symbioses, but fungal penetration into algal cells is uncommon in stratified crustose, foliose and fruticose lichens, which more usually have intraparietal contacts. These may involve substantial thinning of the fungal as well as algal cell walls, which might facilitate more efficient transfer between symbionts. But in other lichens, intraparietal contacts involve peg-like protrusions of the fungal cell wall, which may serve to maintain cell surfaces in contact under the stresses of daily cycles of shrinkage and hydration. By depositing new wall material appositionally or dividing into spores, the algal cell response to fungal penetration may be an essential part of the dynamic equilibrium that permits coordinated histogenesis and maintains what outwardly appears to be a stable collaboration. While the lichen is a superorganism (metaorganism) with emergent properties quite unlike those of its microorganismal constituents, those components and their distinct life histories must be considered as the continuum from which the lichen emerges and to which the dispersing components will later return.

Prospects for and requirements of modern botanical collections in a global context

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Botanical collections have a centuries long tradition in aggregating and curating specimens from plants, fungi, lichens, algal groups and even bacteria. Starting with the European Renaissance period, 15th and 16th centuries, from which a few collections are still extant repositories cover an estimate of several hundred Million of specimens¹ to date.

Drivers for collecting and preserving such collections in early times were frequently bound to strategies of empires to expand spheres of their influence under global perspectives (Fig. 1). Early works like those of Carolus Clusius² (Fig. 2) comprise hundreds of taxa from the region of interest (Iberian Peninsula), but then already including numerous introductions from regions outside Europe. The enormous number of objects flooding Natural History Cabinets in those times necessitated the invention of new methods for suitable classifications ultimately resulting in Systems like Linnaeus' *Systema Naturae*³ to modern times Phylogenetic treatments^{4,5} (Fig. 3). Such systems were and are still utilized to physically arrange collections, storage and retrieval of materials.

Today we face new challenges by an ever-growing number of possible utilizations of herbarium collections⁶. Modern technologies allow for large scale digitization⁷ and thus need innovative approaches to data capture and management⁸, ultimately leading to new approaches for data sharing and publication⁹. These developments force individuals and institutions likewise to respond to challenges for contributing to common goals and elaborate strategies towards a shift of paradigm under the umbrella of innovative Open Science practices²⁰.



Fig. 1. NHM Vienna.



Fig. 2. Frontispiece Clusius 1576.

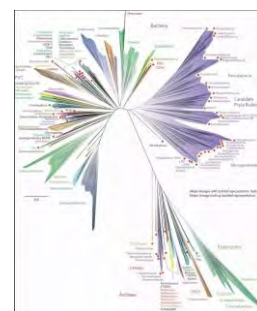


Fig. 3. A new view of the tree of life.

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Antuerpiae, ex officina Christophori Plantini. <https://bibdigital.rjb.csic.es/ident/1/14854>

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10) K. Vohland, S. Eichert, S. Fiedler, M. Kapun, A. Kroh, I. Mehu-Blantar, I. Ott, H. Rainer, M. Schwentner, E. Zimmermann (2022). nhmvienna/OS_Strategy_NHMW: Open Science in Museums v1.0 (v1.0). Zenodo. <https://doi.org/10.5281/zenodo.6505274>

The global re-shuffling of species

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Welcome to the Anthropocene! For the first time in planetary history, one species has become the dominating biophysical force. Consequently, humans are driving the redistribution of biota including plant species at an unprecedented scale, with enormous and rising consequences – for the future of biodiversity and human livelihoods alike. While many plant species are declining and heading towards extinction, other plants are being moved around the globe by humans, essentially changing global biogeography. This human-induced introduction of plant species into new regions have become a defining signature of the Anthropocene.

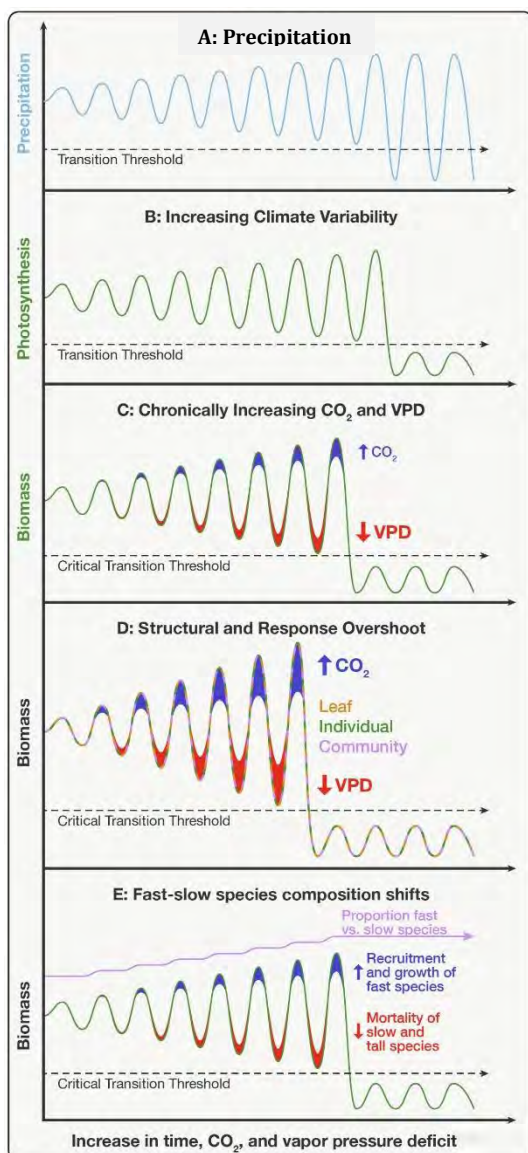
In my talk, I will synthesize key insights into global patterns and drivers of plant invasions. I will highlight likely future trajectories of this phenomenon, present main advances in plant invasion science, and identify new avenues to improve the understanding of alien plant species spread and impacts. Finally, I will provide a perspective on priority questions for biogeography and biodiversity conservation resulting from the global re-shuffling of plant species.

The future of critical transitions at leaf to community scales

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Global ecosystems are increasingly crossing critical transitions (CTs) in their structure and function and, in parallel, are exhibiting decreasing resilience. The mechanisms underlying decreasing resilience and its potential association with CTs are largely unexplored, limiting our predictions of these abrupt transitions and possibly promoting underestimates of future atmospheric CO₂ and temperature. In this presentation I propose and evaluate four mutually-inclusive hypotheses to explain decreasing resilience and links to CTs. The first hypothesis is that climate variability alone drives changing resilience and CTs. The second hypothesis builds on the first, and proposes that rising CO₂ and vapor pressure deficit could promote the appearance of reduced resilience via anomalously high gas exchange during periods of mild climate and anomalously low gas exchange during hot-droughts. Neither hypothesis one or two consider climate-biology interactions. Hypothesis three is that decreasing resilience and CTs result in part from structural overshoot (SO), wherein CO₂-fertilization under mild climate conditions promotes anomalously high biomass that cannot be sustained under hot drought (Hypothesis two) leading to hydraulic failure. The fourth hypothesis is that slower shifts in



community composition towards faster growing and dying species (the fast-slow growth and mortality continuum) promotes decreasing resilience and CTs because of the greater growth under mild climates, and greater mortality during droughts, than slower species.

Hypothesis one has marginal support as resilience has increased and remained stable in boreal and temperate forests, respectively, since 2005, but with no evidence that climate variability had an impact. Tropical regions have exhibited negative resilience since 2005 with climate variability explaining only a small portion of the observations. Hypothesis two is supported by large resilience declines in gross primary production across all biomes, but this impact on photosynthesis did not translate to impacts on wood growth thus is unlikely to be a dominant process of CTs. SO as a cause of resilience declines and CTs is broadly supported (hypothesis three) as is evidence of SO promotion by shifting species compositions. All four of these hypotheses could interact to promote resilience loss and CTs. None of the hypotheses have been adequately tested to draw strong conclusions yet. Nonetheless, there is high-likelihood of globally decreasing resilience and increasing CTs with rising CO₂ and vapor pressure deficit, the latter exacerbating hydraulic constraints. ESMs could represent the mechanisms underlying resilience and SO, mechanisms of overshoot thereby a path exists to improve prediction of tipping points in the climate system.

Fig. 1. The four hypotheses regarding the increasingly common resilience loss and critical transitions in forests. A) precipitation time series exhibiting increasing variance. B) Photosynthesis exhibits increasing variability in concert with precipitation. C) Rising CO₂ and vapor pressure deficit promote the variance, loss of resilience, and CTs. D) Structural overshoot exacerbates the impacts shown in panels B and C. E) Shifts in species composition can further exacerbate the processes in panels A-D.

COMMUNICATIONS

Phylogeography and phylogenomics of the *Hieracium tomentosum* group

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Fig. 1. *H. tomentosum* (photograph of S. Orsenigo)

Hieracium L. s.str. (Asteraceae, Cichorieae) is among the most diverse plant genera in the angiosperms. The extreme morphological variability which characterizes this subgenus is due to complex and not yet understood reproductive strategies combined with extensive interspecific hybridization and polyploidization.

The latest treatment for the Italian flora (1) largely follows Zahn's one (2), dating back to over a century ago and purely based on morphology. Recently, an alternative approach was proposed for some species from the French Alps (3), where also ecology and ploidy level are considered resulting in the recognition of microspecies which are grouped in informal series. Such an approach ought to be applied to Italian territories as well.

This project focuses on *Hieracium tomentosum* L. (Fig. 1) and the numerous taxa supposedly related to it, representing a morphologically distinct group defined by the presence of more or less conspicuously plumose hairs. This group of species is endemic to the W Alps (France, Italy, and Switzerland) extending southwards along the Apennines, with a rather fragmented distribution, up to Abruzzo. Triploids and a single diploid population are known in *H. tomentosum* s.l. thus making it a suitable candidate for studying intra- and inter-specific relationships integrating taxonomy, morphology, ecology, genome size, cytology, and molecular biology.

In particular, we attempted to investigate the phylogeography and phylogenomic of the selected species employing ddRAD-Seq (double digest Restriction-site Associated DNA Sequencing) derived genomic data to infer the relationship of the diploid *H. tomentosum* with the related/derived apomict taxa. The ploidy level of numerous taxa, many of which analyzed for the first time, has been assessed via chromosome counts and flow cytometry. The preliminary phylogenomic results are corroborated by the morphometric and cytometrical analyses and support the separation of the diploid and triploid *H. tomentosum* as distinct taxa with separate ranges. In addition, a series of microspecies with narrow ranges and supposed hybrid origin is recognized.

This project will hopefully result in an alternative new taxonomic treatment for a redefined *H. tomentosum* group. Such an approach could be potentially extended to other *Hieracium* species, thus contributing to shed light on this complex genus.

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Dolines on Karst Plateau as an object of vegetation studies

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Dolines were also one of Professor Poldini's objects of study and he initiated further research into this phenomenon in the region. The article presents some of the latest research results of the research group.

Dolines (also known as sinkholes) are closed depressions in the karst landscape. They are usually sub-circular and funnel-shaped in plan, but can also be elliptical or linear. They have a diameter of a few meters to several meters and are slightly sloping to vertical on the sides. Consequently, their depth generally varies from a few meters to several dozen meters. Different orientation and inclination, which are a result of the terrain undulations, are decisive for the soil and the microclimate. On south-facing slopes, which receive more radiation, the temperature is higher and the relative humidity lower. This leads to differences in the weathering of the rock and differences in the microclimate and soil. This in turn creates the ecological conditions (light, nutrients, humidity, temperature) for plants and plant communities. Dolines are often regarded as microrefugia, i.e. places with special environmental conditions that facilitate the survival of species in the event of environmental changes.

The dolines can be divided into several landscape-vegetation units that are result of different ecological conditions along the slopes. The different ecological conditions are also reflected in the different plant traits (1). There appear also different forest communities in dolines (2). The beginning of the flowering period of plants depend on the ecological conditions along slopes (3).



Figures. The thermal inversion can be well recognized in the spring when snow melts (left), transect along the doline (right).

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2) Jakob, A., Breg Valjavec, M. & Čarni, A. (2025). Determination of forest communities on the basis of small plots (microplots) within the geomorphologically diverse landscape of the Kras plateau (Italy, Slovenia). *Forest Ecosystems*, 12, 100283

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Leaf trait variation across Italian forest endemics: drivers and divergence from widespread congeners

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Despite their biogeographical and conservation relevance, the functional trait space explored by Mediterranean endemic plants still remains largely unknown, particularly for those of forest habitats (1). To what extent these taxa are functionally divergent from related widespread congeners from similar forest habitats is a central question potentially involved into the causes for their restricted distribution, ecological specificity and resource use attitudes. Leaf traits play a critical role in plant growth and survival, largely determining the functional space explored by plants in terms of resource use (2). Analyzing these traits in forest endemic plants and unveiling the range of interspecific variability can thus open a window on their relative position along the acquisition-conservation axis, in the framework of the global leaf economics spectrum (3). Accordingly, we analyzed six core leaf traits (LA, SLA, LMA, LDMC, Nmass, C:N ratio) across 45 endemic taxa (“paleo-“ and “neo-“ endemics) living in Italian forests, either woody or herbaceous, in distant angiosperm and conifer clades. Leaf traits were measured in the field on one representative population per species, mainly in the central, southern and insular regions. The influence of a set of environmental variables and the phylogenetic signal of traits were also examined to identify the major drivers of interspecific variation. Next, we performed species pairs contrasts in 27 pairs formed by the endemic taxon and a closely related widespread congener also of forest habitats, with allopatric, parapatric or sympatric distribution. Overall, trait variation across the endemics was remarkably wide, reflecting their diversity in terms of morpho-functional types, phylogenetic relationships and biogeographical contexts. Endemics were widely distributed along the resource use gradient associated with LA, LMA and Nmass. The herbaceous taxa showed a prevalence of the C and R strategies, while stress-tolerance was prevalent in the woody endemics. Most traits showed a phylogenetic signal based on various metrics used, with Pagel’s λ approaching the Brownian model for LA and LMA. Environmental factors variously influenced trait variation. LA was negatively affected by mean annual temperature and depended on forest type, while LDMC decreased with latitude and precipitation. LMA was positively associated with temperature and varied with ecoregion and forest type, while Nmass decreased with latitude and increased with precipitation. Species pairs analysis pointed to a generally negative effect of the endemic condition on LA, associated with increasing LMA and Nmass. Compared with widespread congeners, this pointed to lower acquisitive ability and stronger resource conservation attitude, also reflected in significant shifts in the CSR strategies. Differences in LA and LMA within allopatric and/or parapatric pairs were overall larger than in sympatric pairs, suggesting the role of vicariance and range separation in the divergence of these traits between related endemic-non endemic taxa of forest habitats.

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2) K.J. Niklas, P. Shi, J. Gielis, J. Schrader, Ü. Niinemets. (2023) Editorial: Leaf functional traits: Ecological and evolutionary implications. *Front. Plant Sci.*, 14

3) J.J. Wright, P.B. Reich, M. Westoby, D.D. Ackerly, Z. Baruch, F. Bongers *et al.* (2004) The worldwide leaf economics spectrum. *Nature*, 428, 821–827

The Italian National Forest Inventory (IFNI2025): a new opportunity for plant diversity monitoring

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Monitoring forest resources through National Forest Inventories (NFIs) is a fundamental tool for policy-making particularly regarding environmental planning and sustainable land management. In Italy, the first NFI dates back to the 1980s followed by a second in 2005 and a third in 2015, each introducing improvements in sampling design and survey protocols to address emerging information needs. The new Italian National Forest Inventory (IFNI2025) represents an innovative program marked by a transition from decennial periodicity to a continuous survey approach with the production of annual estimates. The sampling design involves a first-phase tessellation stratified sampling on a 4 km × 4 km grid followed by a second phase of field surveys on points classified by remote sensing imagery as forest. The second phase sampling will take place annually over a five-year cycle.

For the first time, IFNI2025 will include a systematic survey of variables related to plant and lichen diversity, complementing the assessment of traditional forest attributes. Specifically, data on six plant morpho-functional groups (i.e., terricolous lichens, bryophytes, pteridophytes, forbs, grasses, and woody species) will be collected along transects at second-phase points (approximately 1,300 per year) by personnel from the Carabinieri Command for Forest, Environmental, and Agri-food Protection (CUFAA). These data will be used to estimate the presence/absence and average abundance of the morpho-functional groups at national, regional, and biogeographic scales. The protocol also includes a spatially balanced sampling design covering 10% of the second-phase points with vegetation surveys performed by expert botanists. Diversity indicators will be calculated, including the average number of species (with a focus on alien species and conservation-relevant categories such as specialists and endemics) and their relative abundance. The integration of field data with expert-derived floristic checklists will also be explored to enhance species richness estimates. Custom estimators will be developed to quantify the target parameters and their associated uncertainties. Epiphytic macrolichens will be detected using a simplified method in which an artificial intelligence algorithm is trained to interpret photos of lichens present within a sampling grid. The data will be interpreted based on the main functional traits.

This new protocol represents a significant advancement in national forest monitoring—a major step forward in integrating biodiversity assessments into NFIs—providing consistent and scalable data to support forest conservation and management in Italy.

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2) Hufft, R., Alba, C., & Sahud, A. (2019) Vegetation Monitoring Protocol for measuring and collecting ecological data. *protocols.io*. <https://dx.doi.org/10.17504/protocols.io.4jmguk6>

Plant biodiversity patterns and conservation challenges in Mediterranean minor islands: a comprehensive review

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Mediterranean islands represent one of the world's most diverse archipelagos with ca. 10,000 islands and islets, hosting significant biodiversity components and serving as natural laboratories for testing ecological and evolutionary hypotheses (1). These ecosystems are characterized by high species richness, endemism levels, and unique biotic assemblages, with plant communities showing vulnerability to anthropogenic pressures and global environmental change (2). This review synthesizes current knowledge on biodiversity patterns across Mediterranean minor islands, with special focus on floristic diversity, to identify conservation priorities and research gaps in these vulnerable ecosystems facing increasing threats from human expansion, climate change, and biological invasions. This review was conducted using the MEDIS spatial database, the first comprehensive dataset of Mediterranean islands larger than 0.01 km² (3). We focused specifically on minor islands with area smaller than 50 km², extracting data on geographic variables for 2,217 islands across the Mediterranean Basin. Literature search was performed using Web of Science, Scopus, and Google Scholar databases, combining keywords related to "Mediterranean islands", "biodiversity", "flora", and "conservation". Preliminary results from our analysis identified 588 peer-reviewed studies on Mediterranean minor islands from a comprehensive literature database of 1,411 articles. The geographical distribution shows a clear dominance of Greek (32.3%) and Italian (31.3%) islands, together accounting for 63.6% of all studies, followed by French islands (13.9%, mainly Corsica), Spanish archipelagos (6.8%), Croatian islands (6.0%), Turkish coasts (5.0%), Malta (3.7%), and Cyprus (1.0%). Thematic analysis revealed a slight predominance of faunal studies (47.1%) over floristic research (42.7%), with only 10.2% of studies adopting an integrated approach examining both flora and fauna. Country-specific patterns emerged, with Greece, Italy, and France showing consistent faunal research dominance (53.8%, 51.8%, and 57.9% respectively), while Spain represented an exception with floristic studies prevailing (41.2% vs 35.3%). These preliminary findings demonstrate significant research gaps in Eastern Mediterranean islands (Cyprus, Turkey) and highlight the limited integration between botanical and zoological approaches in Mediterranean island biodiversity studies, with most research focusing on single taxonomic groups rather than ecosystem-level investigations. The observed research bias towards Western Mediterranean islands (Greece, Italy, France) likely reflects greater accessibility, established research infrastructures, and historical scientific traditions, while the predominance of single-taxon studies suggests missed opportunities for understanding ecosystem-level processes and species interactions that are crucial for island conservation strategies. This preliminary review reveals significant geographical and thematic gaps in Mediterranean minor island biodiversity research, emphasizing the urgent need for integrated studies in underrepresented regions, particularly Eastern Mediterranean islands, and for ecosystem-based approaches that simultaneously address flora, fauna, and their ecological interactions to support evidence-based conservation planning.

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Endemism patterns of the vascular flora of Lebanon and ex situ conservation aspects

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Given its high concentration of rare and endangered plant species, Lebanon is recognized as a biodiversity meso-hotspot within the Levant. This study presents a list of vascular plants endemic to Lebanon, their taxonomic diversity, life and chorological forms, spatial distribution across key geomorphological features, comparisons with the floras of Egypt, Iran and Turkey, and conservation status. The list, comprising 168 taxa belonging to 37 families and 97 genera, was compiled through a comprehensive review of published literature, examination of herbarium specimens, and insights from field observations. The five most endemic-rich families are Asteraceae (19%), Lamiaceae (14.3%), Fabaceae (11.3%), Caryophyllaceae (5.6%), and Iridaceae (5.3%). The most endemic-rich genus is *Astragalus* (8.3%), followed by *Centaurea* (4.5%), *Allium* (4.2%), and *Iris* (3%). Hemicryptophytes are the predominant life form (66.8 %), followed by geophytes (13%), and chamaephytes (10%), reflecting the mountainous and Mediterranean character of Lebanon. In terms of spatial distribution, Mount Lebanon and Anti-Lebanon, emerge as centers of endemism, hosting respectively 69 and 20 exclusive taxa. The comparison with other floras highlighted the influences of the neighboring biogeographic regions, mostly Irano-Turanian and Mediterranean. According to IUCN Red List, 53% of the taxa have been assessed, with 9.5% taxa classified as Critically endangered, 28.5% as Endangered, 9.5% as Vulnerable. The checklist is available on an online repository (https://ipt.pensoft.net/resource?r=checklist_of_vascular_plants_endemic_to_lebanon&v=1.0) and is considered dynamic as it will be updated in response to taxonomic modifications driven by genetic analyses and the revision of distribution range. The seeds of 50 taxa were collected and stored in the Sardinian Germplasm Bank (BG-SAR) of the University of Cagliari (Italy), including 14 species strictly endemic to Lebanon, and the others endemic to the Levant. This is the first time that seeds of endemic wild plant species from Lebanon are stored in an international seed bank, paving the way to safeguard these unique genetic resources for future generations, ensuring their preservation even in the face of unforeseen challenges.

Peripheral population or understudied distribution? The tale of *Crepis suffreniana* (Asteraceae) in Italy

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Peripheral populations represent valuable systems for addressing biogeographical and evolutionary hypotheses, often making them key targets for conservation projects. Here, we illustrate the case of *Crepis suffreniana* (DC.) Steud. (Asteraceae), a southern European annual species whose distribution is centered in southern France, reaching eastwards, with a putative disjunct peripheral population, the coastal Migliarino-San Rossore-Massaciuccoli (MSRM hereafter) Regional Park (Tuscany, central Italy; 1). Indeed, the MSRM Regional Park is recognized to host some peripheral populations of widespread European plant species, like the western *Hypericum elodes* L. (Hypericaceae) or the eastern *Symphytum tanaicense* Steven (Boraginaceae), which grow in wet habitats. On the contrary, *C. suffreniana* is known to grow in dryer habitats characterized by sandy soils. Nevertheless, the distribution of this species in Italy is uncertain and understudied, also due to possible confusion with similar taxa (e.g., *C. apula* (Fiori) Babco.). Hence, it is not clear whether the Tuscan population actually represents a peripheral population. Therefore, the main aim of this research is to clarify the actual distribution of *C. suffreniana*. The first step of this investigation was to consult herbarium specimens of *C. suffreniana* and related taxa (*C. apula*, *C. bellidifolia* Loisel., *C. capillaris* (L.) Wallr., *C. neglecta* L. subsp. *neglecta*, *C. neglecta* subsp. *corymbosa* (Ten.) Nyman) in the Herbarium of the University of Pisa (PI). We checked for specimens collected in Italy and Southern France, to test the reliability of the diagnostic features proposed in previous works (1,2,3). Some of the diacritical characters, like the number of involucre bracts, was not useful in the morphological distinction of the taxa. On the other hand, we observed that the beak of the cypsela in *C. suffreniana* (Figure 1), together with the annual life form, the relatively small flowers, and the flower heads nodding before anthesis, allow to clearly distinguish this species from related ones. Then, at the end of April 2025, we conducted some field surveys in the MSRM Regional Park, to look for this species. We found *C. suffreniana* at many sites in the central portion of the MSRM Regional Park (Figure 1), growing in subpopulations with more than 50 plants per hectare. We observed that *C. suffreniana* preferably grows in open dry meadows dominated by therophytes, although some individual plants were found in small canopy openings of the holm oak forest. Interestingly, *C. suffreniana* can grow together with other related taxa like *C. capillaris* and *C. neglecta* subsp. *neglecta*, although these latter species preferably grow in slightly wetter microhabitats. These preliminary investigations confirm the occurrence of *C. suffreniana* in the MSRM Regional Park, highlighting the role of this protected area in preserving drylands and the species linked to these habitats. A critical revision of specimens from European herbaria is currently ongoing to understand the actual distribution of *C. suffreniana*. We are also collecting fruits from Tuscany and France for *ex situ* conservation and to check whether the chromosome number and/or karyotype structure differ between core and peripheral populations, since karyotype alterations are known to be an important driver of diversification in the genus *Crepis*.



Fig. 1. On the left, *Crepis suffreniana* growing in the MSRM Regional Park. On the right, a close-up of the cypsela

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Leaf spectral properties and leaf traits of four alpine species of butterworts (*Pinguicula* L.)

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Understanding the traits that influence the responses of species to climate change is a crucial aspect of predicting future community composition and ecosystem function. Leaf functional traits are informative data to understand adaptation and acclimatization of species to ecological niches. Moreover, different plant species could have unique spectral reflectance signatures that might provide information to recognize and discriminate plant species.

We characterized leaf morpho-functional traits of four species of *Pinguicula* with different ecological niches, from dump meadows to rocky walls. Specifically, we studied *P. alpina*, *P. leptoceras*, *P. vulgaris* and *P. poldinii*. Sampling activities took place in the Dolomites, north-eastern side of European Alpine Arc. We measured the following environmental parameters: temperature, relative humidity, light intensity, light spectrum. As leaf functional traits we considered leaf dry matter content and specific leaf area. In order to characterize the photosynthetic adaptation in alpine butterworts, we quantified leaf pigment contents and we measured photosynthesis *in-situ* and leaf reflectance (400-1000 nm). Also, we observed leaf surface features, trichomes and glands morphology via scanning electron microscopy (SEM).

Despite a lower concentration of chlorophylls, *P. poldinii* and *P. alpina* showed a higher photosynthetic efficiency (e.g. yield of open photosystem II reaction centres in light) compared to the other two species. Since the different species showed different leaf reflectance spectra, our findings corroborate the ongoing hypothesis that leaf reflectance can be a useful tool to interpret data related to leaf traits, pigment composition and tissues organization.

By providing a comprehensive scenario of leaf morpho-functional characteristics and leaf spectra signature, we show that the studied-species are coherently optimized to their prevailing environmental conditions.

Ngezi-Vumawimbi Forest Reserve, Pemba Island, Zanzibar (Tanzania): a unique biogeographical hotspot

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The Ngezi-Vumawimbi Forest Reserve is the largest last remnant of natural vegetation covering Pemba Island, Zanzibar, Tanzania. The most comprehensive biodiversity report of this area is based on Beentje field work (1). The gazetted area of the Forest Reserve covers around 20 km², including moist forest and a giant heath vegetation dominated by *Erica mafiensis* (Engl.) Dorr. Other relevant vegetation types are coastal evergreen thicket, dry coastal forest, mangrove, wetlands and secondary bush, resulting from over harvesting of the forest. According to Beentje (1) Ngezi Forest is unique at global level since it shows an assemblage of species that is not paralleled in any other East African forest. It is, in fact, the only forest in East Africa where species restricted to coastal East African forests [e.g. *Rhodognaphalon mossambicense* (A.Robyns) A.Robyns], afromontane elements [*Ensete ventricosum* (Welw.) Cheesman, *Erica Tourn. ex L.*, *Cassipourea gummiflua* Tul.], Eastern Indian species [e.g. *Samadera indica* Gaertn., *Donella lanceolata* (Blume) Aubrév.], Guineo-Congolian forest species [*Uapaca guineensis* Müll.Arg.] and Mascarene and Madagascar species [e.g. *Chrysalidocarpus* H.Wendl., *Typhonodorum lindleyanum* Schott] occur together.

In December 2024, we realized a biodiversity baseline survey in the Forest Reserve in the framework of the GREEN Project (Generating Resilience through Ecological Enhancement in Ngezi-Vumawimbi Forest), led by Istituto Oikos, by conducting floristic and forestry surveys. Botanical surveys preliminary data, performed in all the main habitats of the Forest Reserve, reveals hidden and extraordinary biodiversity. We found at least 325 plant species, of which approximately 20% represent species never reported for Ngezi-Vumawimbi Forest Reserve or for the Island of Pemba.

We confirmed the presence of species known only for few localities in East Africa (e.g. *Allophylus vestitus* F.G.Davies, *Strophanthus zimmermannianus* Monach.) or endemic of Pemba (e.g. *Chrysalidocarpus pembanus* H.E.Moore) listed as Endangered, Vulnerable or Near Threatened at global level in IUCN Red List. Moreover, we confirm the presence *Intsia bijuga* (Colebr.) Kuntze, an important timber tree distributed primarily in Southeast Asia and Madagascar, considered as Near Threatened at global level. This species, only known for Pemba from old herbarium specimens and no longer confirmed recently, dominate the evergreen coastal forest in Vumawimbi, and represent the only population of *I. bijuga* in continental Africa. This finding has considerable biogeographical and conservation implications. The *Intsia* forest at Vumawimbi forms a well-structured and ecologically distinctive forest type in which the species is locally abundant, and often dominant. This forest supports and defines a distinct assemblage of plant species not found elsewhere in Africa, with at least two species of orchids considered new two sciences (*Disperis* sp. nov. and *Bulbophyllum* sp. nov.) currently under study.

Despite these exciting discoveries, the conservation status of this highly diverse Forest Reserve is critical. The timber trees are continuously overexploited through illegal logging. The *Erica mafiensis* heathland is annually crossed by fires that threaten this unique ecosystem. The whole area of Vumawimbi dominated by *Intsia* forest (3 × 1 km), one of the richest areas of the Reserve, is designated for the development of a new 7-stars “eco-resort”. We aim to shed light on the biodiversity and biogeographical importance Ngezi-Vumawimbi Nature Forest Reserve, a critical contributor to the overall importance of the Zanzibar-Inhambane coastal forest hotspot of biodiversity (2), enabling conservation partners, public institutions and investors to adopt the most suitable management practices to guarantee its long-term protection.

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Diachronic analysis of Etna high-mountain plant communities

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Understanding the vegetation dynamics in the high-mountain habitats is crucial for assessing the long-term effects of natural disturbances and climate variability (1). The use of historical maps is a powerful tool in the analysis of landscape dynamics, offering a unique combination of temporal and spatial information that historical documents alone often lack (2). In this study, a diachronic analysis of the high-altitude plant communities of Mt. Etna (Sicily) was conducted over a 60-year timespan by comparing four vegetation maps dated 1965, 1978, 2000 (3), and 2025. The study area ranges from approximately 1,400 m a.s.l. up to the peak of the volcano (3,403 m a.s.l.), encompassing 14 vegetation types consistently represented in all maps.

All maps were georeferenced and digitized using a uniform spatial framework in QGIS. The validation of each vegetation polygon was made through literature data, field surveys, and photointerpretation. Temporal changes were assessed by calculating absolute and percentage area variations, Shannon diversity (H') and Pielou's evenness (J') indices. Statistical significance of differences among years was tested using the Kruskal-Wallis test. A turnover index was also applied to evaluate compositional shifts between consecutive years.

Results indicate substantial transformations, with pioneer communities such as the *Senecioni aetnensis-Anthemidetum aetnensis* and the *Astragaletum siculi* showing marked regressions, likely due to successional dynamics and increasing vegetation cover on formerly bare substrates. In contrast, woody communities like those physiognomically featured by *Juniperus hemisphaerica*, *Fagus sylvatica* and *Pinus nigra* subsp. *calabrica* expanded significantly, suggesting an upward shift of forest belts. Moreover, although *Betula etnensis* experienced a slight decline between 2000 and 2025, it underwent a considerable expansion between 1965 and 2025, with an overall increase of 216.3%, likely driven by its pioneer character and the disturbance dynamics of the area. Diversity and evenness values fluctuated, reflecting changing complexity in landscape structure. Turnover analysis highlighted intense transitions particularly between 1978 and 2000.

This long-term study provides valuable insight into the ongoing ecological evolution of Mt. Etna landscape and offers a scientifically based approach for future monitoring and conservation actions.

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Floristic and Ecological Characterization of Endemic Plants in Northern Italy: spatial patterns and environmental drivers

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Floristic cartography aims to produce distribution maps of vascular plants based on a grid system (i.e. presence/absence), by systematically surveying a territory without prioritizing specific taxa or habitats. This approach allows for an overall and comparable estimate of floristic biodiversity across the various geographical areas under study. The chorological study of the Italian flora is especially useful in identifying distribution patterns along north-south and east-west axes, which are linked to the country's biogeographic history and geo-ecological complexity.

Distribution maps are also essential tools for compiling lists of species to be protected (e.g., legislative annexes, red lists, etc.), including rare or endemic species, and for identifying pathways of introduction of alien species (neophytes), highlighting their expansion trends and level of invasiveness. Unlike in some Central European countries, floristic cartography in Italy developed more recently, starting in the 1970s with numerous local-scale projects.

Focusing on Northern Italy, over the past three decades, various provincial and regional projects have recorded more than 6,000,000 floristic data points, corresponding to over 1,600,000 records when grouped by quadrats. In recent decades, there have been several publications of distributional nature at various scales, but the most comprehensive work, in terms of unified territory covered, is undoubtedly the recent *Atlas of Endemic Flora in Northern Italy*. This includes distribution maps by MTB grid cells (i.e., quadrats) of 447 northern Italy endemic entities over a territory spanning 121,974 km² (corresponding to 3,612 grid cells), encompassing eight Italian regions (Liguria, Piedmont, Aosta Valley, Lombardy, Trentino-Alto Adige, Veneto, Friuli Venezia Giulia, Emilia-Romagna).

Based on this dataset - composed of 32,939 floristic records (from field surveys, herbaria, and bibliographic sources) attributed to grid cells and sourced from the work of 19 local floristic cartography projects - the present study aims to analyze the spatial distribution of floristic diversity of endemic species in Northern Italy, with a focus on the main factors shaping its current structure.

Specifically, after analyzing the flora endemic to Northern Italy from the perspective of biological spectrum and ecological preference using geospatial tools, this study compares the effects of past environmental drivers (e.g., glacial extent, past climate, past forest cover, geomorphology) with those of present-day drivers (current climate, land use and land cover), in order to assess the most significant predictors of floristic biodiversity in the specific context of Northern Italy.

This is a highly heterogeneous area, with 46% of grid cells falling within the Alpine zone, 34% in the Venetian-Po Plain, and 20% belonging to the Northern Apennines. Accurate collection of floristic data is an essential prerequisite for understanding biodiversity and implementing effective species conservation strategies, especially for endemics, allowing for informed land management, sustainable soil use, and planning in the context of climate change and potential future extinctions.

Ellenberg Indicator Values demonstrate community shift and habitat loss in the forest of San Rossore (Nature 2000 Site ‘Selva Pisana’)

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The Nature 2000 Site ‘Selva Pisana’ (Parco Regionale di Migliarino, San Rossore, Massaciuccoli) includes three habitats distributed in a continuum: riparian forests (91F0 Habitat code), alluvial forests (91E0), and small patches of sub-Atlantic and medio-European forests (9160).

Detailed floristic and vegetation inventories are available for this site from the second part of the last century (1,2,3). According to them, riparian forests can be attributed to the association *Fraxino-Quercetum roboris* Gellini, Pedrotti and Venanzoni 1986, with the subass. *carpinetosum betuli*, and alluvial forests to the association *Carici-Fraxinetum angustifoliae* Pedrotti, 1970, with the subass. *alnetosum* Gellini, Pedrotti and Venanzoni 1986. Around year 2000, the former association was still the most diffused broadleaved vegetation in the area, while the latter was restricted to very humid soils, often flooded also in spring and summer (2,3). In the last two decades, treefall increased dramatically, with *Quercus robur* being the most affected species. Trees underwent both trunk crash and uprooting, which was generally ascribed to extreme summer drought (Fig. 1). To confirm this hypothesis, historical data were compared with floristic inventories carried out in the corresponding forest stands from 2023 to 2025. The Ecological Indicator Values of Ellenberg (EIV) were attributed to species according to the adaptation to the Italian flora made by Pignatti and were weighted according with the ordinal method transforming the Braun-Blanquet cover values in figures from 1 to 9.

The analysis of vegetation in a stand which was once attributed to the association *Fraxino-Quercetum roboris* with an abundant layer of *Ruscus aculeatus* (1,3), showed today a marked shift towards species indicating increased soil humidity and higher solar radiation (Fig. 2). The occurrence of hygrophilous species like *Limniris pseudacorus* and *Carex riparia* in the understory, so as the rare presence of *Quercus robur* in the tree



Fig. 1. Up-rooted tree of *Quercus robur* in the broadleaved forest of San Rossore

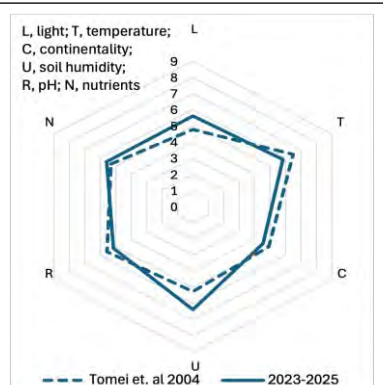


Fig. 2. Changes in the Ecological Indicator Values of Ellenberg for the understory vegetation of a meso-hygrophilous forest stand in the last 20 years

layer, indicate a shift of vegetation towards the association *Carici-Fraxinetum angustifoliae*. These results support the hypothesis that the primary cause of tree dieback is increased soil humidity rather than drought, which was probably caused by the abandonment of drainage-channel management. The expansion of alluvial forests to the expense of more mesophile riparian forests is occurring in several parts of the forest of San Rossore and should deserve special attention. Indeed, riparian forests are High Conservation Value habitats in the Mediterranean region and host extra-zonal species like *Quercus robur*,

Carpinus betulus, *Veronica montana*, *Cardamine apennina*, *Digitalis micrantha*, and *Lilium bulbiferum*, which cannot ‘move’ to drier soils following natural dynamics, because these soils are occupied by farmland, and industrial and urban settlements.

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The mycobiome of declining holm-oak populations of the Maremma Regional Park (Italy): Insight into fungal community dynamics and potential drivers of tree decline

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Holm oak (*Quercus ilex* L.) is a key element of the ecological and socio-economic services of forest ecosystems in the Mediterranean area, but over the last few decades extensive dieback and mortality episodes have been documented on this species after severe and recurrent drought events.

The main aim of SpecFor project (PRIN 2022) is to provide forest managers with the necessary tools to monitor and detect early signs of tree decline, and to implement effective management strategies to mitigate the negative impacts of biotic stresses on holm oak populations. By combining advanced remote sensing techniques with traditional field survey methods and molecular analyses, the project aims at developing a comprehensive approach to understanding the complex interactions between environmental factors, tree health, and fungal communities in Mediterranean forests. Within the Maremma Regional Park (Italy)

(Fig. 1), two areas were selected, characterized respectively by an Elevated (ED) and low (LD) degree of decline. In each area, 10 “healthy” and 10 “declining” trees were sampled based on crown conditions. Protection regulations have ensured the absence of anthropogenic disturbance for at least 30 years in the selected areas. Using Next Generation Sequencing (NGS) metabarcoding, we investigated the ecological role of canopy fungal communities in both ED and LD areas. Fungal community composition was analysed by sequencing the ITS2 region using Illumina MiSeq technology; the functional traits were assigned by the tool FUNGuild. Metagenomic analyses enabled us to characterize the fungal communities associated with declining and low or non-declining holm oak trees in both areas. A statistically significant difference was observed in beta diversity, although not in alpha diversity (Fig.2). Differences in the relative abundance of taxonomic groups between the two areas revealed that the most abundant family in the LD area was Gnomoniaceae, whereas Myriangiaceae, Botryosphaeriaceae, Celotheliaceae, and Trichomeriaceae were more abundant in the high-decline area (ED). The functional analysis revealed a higher prevalence of microorganisms classified as pathogens in the ED area, whereas saprotrophs were more prevalent in the LD area. These results suggest that changes in fungal community composition—particularly the increased prevalence of pathogenic taxa—may play a significant role in holm oak decline.

By highlighting key fungal indicators of tree health, this study contributes to a deeper understanding of the biotic factors involved in forest decline and supports the development of targeted monitoring and management strategies for the conservation of Mediterranean holm oak ecosystems.



Fig. 1. Holm oak stands in the ED area

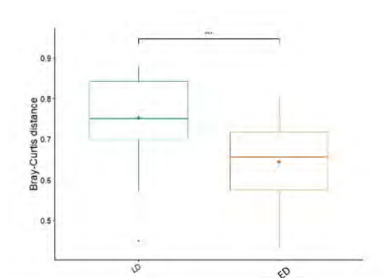


Fig. 2. Beta diversity index based on Bray-Curtis coefficient

Influence of the management of alpine Norway spruce forests on plant and soil mycobiomes

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Forest ecosystem biodiversity is strongly influenced by anthropogenic disturbances, including silvicultural practices that range from intensive management aimed at maximizing ecosystem services to interventions supporting renaturalisation (1). In this context, environmental DNA analysis is a valuable tool for effective biomonitoring and biodiversity assessment, particularly in relation to ecosystem responses to disturbance (2). The plant microbiome comprises a wide variety of microorganisms, including fungal endophytes, which can colonize internal plant tissues and influence host health and development, enhancing plant resilience to both abiotic and biotic stress (3). In particular, fungal endophytes represent promising candidates as bioindicators for evaluating plant health and forest ecosystem resilience under different management or renaturalisation regimes (4).

The aim of this work was to compare mycobiome diversity in soil and tree tissue between two alpine spruce stands in the Adamello-Brenta Nature Park (Italian Alps), one intensively managed through thinning and the other unmanaged for over 40 years. The objective was to assess how forest management may influence fungal community structure.

In each area, five mature Norway spruce trees were selected based on comparable size and sociological position within the stand. From each tree, samples of twigs, wood, roots, and a representative bulk soil sample were collected for ITS-based fungal community analysis. The ITS2 region was amplified and sequenced using HT-NGS. Bioinformatic processing of the sequencing data was performed using QIIME2 and the statistical analyses were conducted in the R environment.

The results show that in soil samples from the managed forest, *Cortinarius*, *Oidiodendron*, and *Tomentella* were dominant, while *Helotiales* and *Microascales* were most abundant in plant tissues. In the unmanaged forest, soil was enriched in *Cortinarius*, *Elaphomyces*, and other *Agaricomycetes*, with *Lachnum* frequently occurring in plant tissues. Moreover, Beta-diversity, core microbiome, taxa composition and relative abundances were different in both tree tissues and soil in the two areas along with the ecological guilds.

In conclusion, forest management significantly influences the structure of fungal communities in both soil and tree compartments of Alpine spruce forests. The observed taxonomic and functional differences suggest that specific fungal taxa may serve as sensitive biomarkers of management regimes and ecosystem health and may become good candidates to evaluate the efficacy of forest management in the future.

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Multiscale assessment of understory functional diversity components and their relationships with forest structure and canopy cover

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The understory accounts for the majority of the diversity in temperate forests (Gilliam, 2014). Functional diversity is an especially relevant indicator, and previous studies have shown how it relates to forest age and management types (Chelli et al., 2024; Chianucci et al., 2024).

We studied 28 beech forest sites in a National Park in Italy (Foreste Casentinesi, Monte Falterona e Campigna) that features coppice, high forest and unmanaged old-growth stands. We apply a novel multiscale approach based on Rao's Quadratic Entropy. We considered functional diversity at stand scale (γ), in small-scale subunits (α), between subunits (β), as well as decomposing the latter in terms of redundancy, uniqueness and clustering. Forest structural features and canopy cover have been quantified in each stand with terrestrial laser scanning and digital cover photography and related to the understory functional diversity at each scale through the use of functional linear models.

We show that stand-scale higher median tree height, but not canopy cover, is related to an increase in functional diversity at small scales, together with an increase in redundancy and functional clustering, while at stand scale there is no evidence of a change in functional diversity and redundancy.

Our findings have implications for forest management and monitoring. First, we highlight the importance of median tree height, which is often overlooked in favour of canopy closure when studying relationships with understory functional diversity. Second, the employed sampling design enabled the identification of relationships at particular spatial and ecological scales that might have been overlooked with the classic stand-level vegetation survey.

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Assessment of landscape, structural and biodiversity metrics of afforestation actions over the last 50 years in the Milan Metropolitan area

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Afforestation actions have been performed from the '70s across the urban and peri urban areas of Milan especially linked to the net of protected areas. The performances of such actions have never or rarely been assessed to understand their success in comparison to more natural forest stands. The aim of our work is to evaluate the success of some significant afforestation areas (AA) carried out in the Milan Metropolitan area over the last 50 years with respect to landscape, woodland structure/composition and biodiversity metrics. Such woodlands will be compared to old woodland stands (OWSs) and woodlands deriving from spontaneous reforestation (SR). Specifically, to assess the evolution of landscape structure and related processes, we conducted a historical landscape analysis of Land Use and Land Cover (LULC) (1) comparing a historical LULC map retrieved from the Second Military Survey of the Habsburg Empire (1818–1829) with a current one from 2021. To detect the composition and structure of 58 AA in comparison to 9 OWS and 11 SR, we performed vegetation plots of 10 × 10 m collecting data on the stand age, species presence and cover as well as we measured height and diameter of trees. Plots have been analysed with cluster and CCA analyses. Parallely, we surveyed with Zeb Horizon (GeoSLAM Ltd., Nottingham, UK) mobile laser scanner (MLS) more than 30 circular plots with a radius of 13 metres to characterise the structure of the different afforestation measures (e.g. DBH, tree height, vertical and horizontal forest structural diversity metrics). The analysis was conducted using the forestry module of the LiDAR360 software (GreenValley International, Berkeley, CA, USA).

Preliminary results showed that the prevailing trend within the whole study area is characterized by a marked decline in agricultural lands, primarily driven by urban expansion. Notably, the existing protected woodland areas - namely, in most cases, woodland remnants recorded since 1820, such as the Bosco di Cusago (Fig. 1) - have also experienced substantial reduction, mainly due to the encroachment of agricultural activities. Cluster analysis and CCA identified main clusters according to species composition, and tree cover and type and age of the stands. As expected OWS and SR formed distinct branches with respect to young (less than 20 years) and old (more than 20 years) forest stands. These last were only partially differentiated. Analyses about woodland structure showed that the distribution of measured structural parameters is statistically significant among the various types of afforestation (Fig. 2), exploiting the potential of these new technologies to complement and extend classical ground surveys to a larger spatial and temporal scale. In conclusion, findings highlight the value of integrating historical, field-based, and remote sensing methods to assess afforestation outcomes and guide future planning in the greening of urban areas (2).



Fig. 1. The old forest of Cusago (MI)

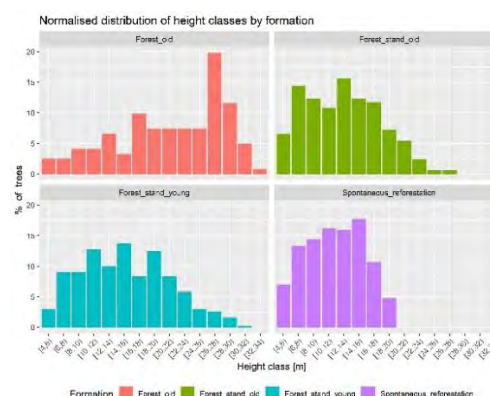


Fig. 2. Number of trees of each class grouped by forest type

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Linking forest structure and plant diversity across secondary forest succession

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Forests cover approximately 30% of the Earth's surface and host most of the global terrestrial biodiversity, which underpins a wide range of essential ecosystem services. Monitoring forest biodiversity is therefore crucial for developing and implementing effective conservation strategies. A key driver of biodiversity at local scale is the forest structure, both in its vertical and horizontal components, which influence species occurrence and distribution by altering microclimatic conditions and determining the availability of resources and ecological niches. Forest structure, in turn, is shaped by multiple factors, including management practices, disturbance regimes, and forest age or successional stage.

In Europe, the widespread abandonment of agricultural and pastoral practices has led to the spontaneous expansion of forests, offering an opportunity to investigate how forest structure develops over time and how it affects plant diversity. During secondary succession, both vertical and horizontal forest structures undergo significant changes, which are expected to influence the diversity of the understorey plant community, from both taxonomic and functional perspectives.

Remote sensing is recognised as a key instrument for monitoring ecosystems and biodiversity. In particular, LiDAR has great potential for characterizing forest structural attributes, as it enables the reconstruction of the three-dimensional shape of vegetation and terrain. Unmanned Laser Scanning (ULS) offers an effective trade-off between high-resolution data acquisition and the capacity to survey large and inaccessible areas (1).

In this study, we investigate how forest structural indices derived from LiDAR data relate to different components of plant diversity along a secondary successional gradient, using a space-for-time approach. Historical orthophotos from the past 70 years (1954, 1978, 2000, 2020) were used to identify and date patches that transitioned from meadows to forest at different times, allowing us to define four successional stages in two Italian regions: Friuli Venezia Giulia and Tuscany. In each region, we analysed four chronosequences, each composed of four stages, for a total of 32 survey plots of 531 m². Vegetation surveys were conducted in three 5 m × 5 m subplots per plot, estimating the percentage cover of all vascular plant species, categorized into herbaceous plants, shrubs, and trees. Species richness and Shannon diversity index were calculated, along with functional diversity indices based on traits retrieved from the literature.

LiDAR data were acquired during the leaf-off season using a Zenmuse L1 sensor mounted on a DJI Matrice 300. The resulting point clouds, reaching 1,500 points/m², provide a substantially higher resolution than most studies in this field. These data allowed for the extraction of highly detailed structural metrics and a more precise characterization of forest architecture. Point clouds were denoised, normalized, and cropped to each 531 m² plot using the R environment. To describe forest structure, we calculated several metrics related to height heterogeneity (e.g., standard deviation, skewness, Gini index of vegetation heights), canopy gaps (number, area, and percentage of gaps at different heights), and the vertical forest profile (e.g., the density of returns at varying heights). Among the tested metrics, gap-related metrics showed the greater association with richness of total and herbaceous species both in Friuli and in Tuscany.

Understanding these dynamics is essential for interpreting biodiversity changes in expanding forest ecosystems and holds great potential for mapping biodiversity in remote forests based on their structural attributes.

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Shifts in understory diversity and thermophilization under climate change are modulated by canopy buffering: evidence from a 5-years re-survey study across European forests

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The herbaceous layer accounts for over 80% of vascular plant diversity in temperate forests and plays a critical role in key ecosystem functions, including productivity, nutrient cycling, and energy transfer (1). Understory communities are particularly sensitive to environmental changes, with climate change currently being one of the most pressing threats. This affects understory communities both directly—by altering temperature and moisture regimes at the forest floor, thereby limiting plant growth and functioning—and indirectly, through its influence on forest dynamics (2). However, these impacts partly depend on overstory structure, canopy density and forest continuity, that can provide more or less microclimatic buffering. Understanding the role of forest structure on the responses of understory communities to disturbance and climate change is therefore essential for predicting biodiversity trajectories under global change and identifying the best management practices to mitigate the impacts (3).

In this study, we investigated the five-year temporal shifts (2018–2023) in three facets of understory diversity, taxonomic (species richness and Shannon diversity), functional (functional richness and functional evenness), and phylogenetic (phylogenetic diversity and phylogenetic dispersion), in relation to forest structure and climate variability across European forests. This period was marked by recurrent and intense heatwaves and droughts, offering the opportunity to detect possible early signals of climate-driven community change at the continental scale. We resurveyed 216 semi-permanent plots across 45 temperate broadleaf forests initially surveyed in 2018. The plots were distributed along a latitudinal gradient from Italy to Norway, spanning three forest density levels—open, intermediate, and dense—as well as a gradient from the forest edge to the forest core. This sampling design enabled us to assess how forest structure and position (distance from the forest edge) modulate understory responses to macroclimatic variation via microclimatic buffering.

Overall, we observed consistent directional shifts toward less diverse and more thermophilous understory communities in 2023, as assessed using linear mixed models. However, the magnitude of these shifts varied across density levels, highlighting the importance of forest type. Remarkably, denser canopies strongly mitigated the degree of thermophilization and buffered the decline in various diversity indices, although such effects were not always consistent between the three facets examined (taxonomic, functional, phylogenetic). These findings show the importance of structural complexity and stand density in moderating the impacts of global warming on understory communities, and thus the need to consider forest structure when assessing biodiversity responses to climate change.

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When *Trebouxia* swims: flagellate cells and hidden sex in the life cycle of a lichen photobiont

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Trebouxia is a genus of green microalgae that are the most common photobionts of lichens, but can also occur as free-living organisms in some aeroterrestrial environments. Despite their ecological importance and many years of research, key aspects of the life cycle of this genus remain poorly understood, in particular the role of flagellate cells.

In this study we applied a novel light microscopy approach that allows continuous observation and video documentation of flagellate cells release and development in axenic cultures of four *Trebouxia* species (*T. decolorans*, *T. vaga*, *T. gelatinosa*, and *T. angustilobata*), representing three of the four major phylogenetic clades of the genus.

Using this technique, we identified two different morphotypes of zoospores (elongate vs. subspherical) consistently present across all species studied. These observations were confirmed in detail by scanning electron microscopy (SEM). Remarkably, the morphological dimorphism of flagellate cells was already described almost a century ago by Otto Jaag (1) in a single *Trebouxia* species, but this information was lost in all subsequent literature.

Occasional fusion events, exclusively between elongate flagellate cells, were observed and documented, leading to the formation of tetraflagellate cells, which we interpreted as planozygotes and eventually zygotes (Fig. 1). SEM images documented the stages of the plasmogamic process with unprecedented clarity (Fig. 2). Using our light microscopy method, we were able to follow the development of a zygote over several days and observed a division of the chloroplast into four parts after ten days, which corresponds to the beginning of a meiotic event.

Molecular analyses confirmed the presence of key meiotic genes in the genomes of eight *Trebouxia* species and their expression in the transcriptome of *T. gelatinosa*, providing strong evidence that *Trebouxia* is capable of active sexual reproduction.

These results revise our understanding of the biology of *Trebouxia* by revealing a sexual cycle in a genus long thought to be predominantly asexual, which has far-reaching implications for the biology of lichen symbiosis. The importance of studying the ploidy of the flagellate cells for a deeper understanding of the life cycle of this organism is also discussed.

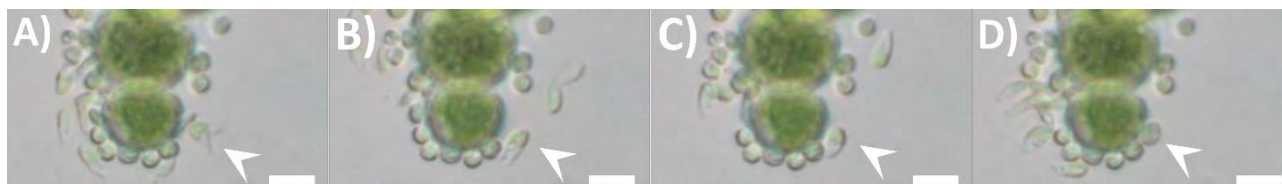


Fig. 1A-1D. Plasmogamic steps of two flagellate cells of *Trebouxia angustilobata* at the light microscope

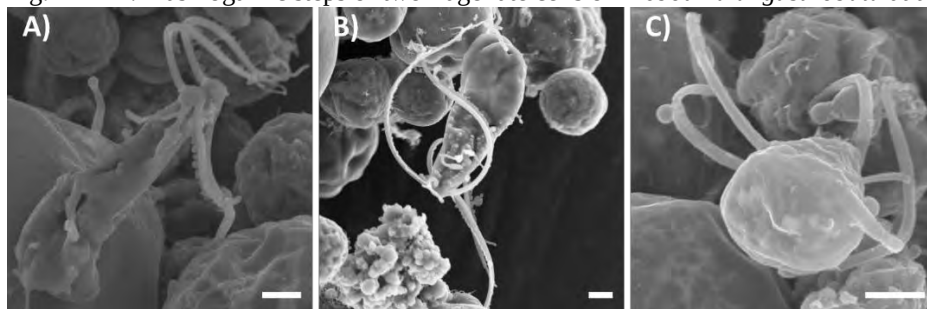


Fig. 2A-2C. SEM photomicrographs of elongate flagellate cells of *Trebouxia angustilobata* undergoing cell fusion

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Detailed floral traits widen the discussion on pollination strategy of *Papaver rhoeas* L. (Papaveraceae)

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The red poppy, *Papaver rhoeas*, is a well-known and common species originated in the East Mediterranean and most likely disseminated along the coast of the Mediterranean around 8000 years ago, backed by the expansion of farming practices as an agricultural weed. Further spread into Central Europe is reported to have occurred approximately 5000 years ago.

Papaver rhoeas is a self-incompatible species and several studies have deepened how its gametophytic self-incompatibility system operates. Consequently, the species relies on pollinators for reproduction.

Floral traits have been largely investigated and analysed to define their role as attractant for pollinators. In East Mediterranean glaphyrids beetles (Glaphyridae: Coleoptera) are reported as main pollinators while in Central European social and solitary bees are the main pollinators, together with flies and beetles. Red poppy flowers are large, bowl-shaped, radially symmetrical, with a dark centre created by the stamen. They are nectarless and result scentless and red to human senses. Most of the debates on floral biology of red poppy involved the interaction between floral traits and insects, mainly focusing on their receptor-based colour vision considering that insect visual spectrum includes also ultraviolet (UV) wavelengths.

First studies on floral reflectance reported that flowers of Eastern Mediterranean poppies reflect exclusively in the red part of the spectrum, while those of Central European poppies reflect both red and ultraviolet (UV) light (1). This was interpreted as a convergence with beetle pollination in Eastern Mediterranean areas and with other insects in Central Europe. More recently, the hypothesis that geographical spread of *P. rhoeas* caused modifications of the colour of its flowers was further investigated and discussed considering the spectral sensitivities of the photoreceptors of the glaphyrids beetles and the honeybees (2). The glaphyrid beetle *Pygopleurus israelitus* has receptors maximally sensitive to UV, green, and red. Differently, most bees have a colour vision based on receptors maximally sensitive not to red but to UV, blue, and green. As for flower reflectance, flowers of Eastern Mediterranean poppies reflect exclusively in the red part of the spectrum, those of Central European poppies reflect both red and UV light. Therefore, results proved that the shift from pollination by glaphyrid beetles (Glaphyridae) to bees occurred alongside changes in flower reflectance.

Our study was carried out in Italy, more specifically in several sites around the cities of Parma and Naples. We focused our attention on several morphological and functional floral traits and associated them not only with the types of insects but also with their behaviour while approaching and visiting the flower. We focused our attention on the dark centre of the corolla especially considering the dark spots often occurring at the base of the petals. Results showed that several types of spots can occur on the petals and that spots, when present, differ in shape, size and colour. Insect visiting the flowers were beetles, bees and hoverflies and the role of different types of petal spots on insect attraction was highlighted. Moreover, we also analysed the flower longevity showing that flowers do not last one day only as previously reported, and that petal orientation changed throughout the day. Such specific flower functional trait was associated to the behaviour of the insects visiting the flower at different time of the day suggesting that the insect interest to the red poppy flowers is not limited to pollen feeding.

Overall, we highlighted that the observation of detailed floral traits remains often overlooked and this occurs even in well-known and widely distributed species as the red poppy. The role of floral traits as petal spots and petal orientation is generally accepted as associated to the interaction with pollinators. Nevertheless, to shed light on the progressive evolution of flower floral specific traits, research on flower-pollinator interactions must always be included. Such studies should not be limited to the list of insect species but must include pollinator behaviour during the flower visits.

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Plant-plant and plant-animal interactions in the recruitment of *Juniperus turbinata* Guss. in Sardinian rocky coastal habitats

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The positive interactions, both among plants and between plants and animals, are crucial for ecosystem functioning: in particular, these are fundamental for maintaining biological diversity, specifically in stressful environments (1, 2). Facilitative interactions are vital not only for individual species recruitment but also for shaping the overall structure of plant communities (2). However, there is a lack of knowledge on the effects of stress gradients on plant-plant facilitation in terrestrial ecosystems.

This research investigates the complex plant-plant and plant-animal interactions involved in the recruitment of *Juniperus turbinata* Guss. in Sardinian rocky coastal environments. The study uses two manipulative experiments to address two key questions: 1) how the survival of juvenile junipers is influenced by the salt gradient and microhabitat, and/or a combination of these factors; and 2) how seed germination and the survival of juniper seedlings are influenced by seed origin, microhabitat, and/or interactions among these factors.

The first experiment was conducted at La Frana (Sassari, North-Western Sardinia). The study site was divided into three homogeneous sub-areas, and each of them was divided into three belts, following the soil salinity gradient already measured (3): starting from the shoreline 0-50 m (F1), 51-100 m (F2), and 101-150 m (F3). Overall, 180 juvenile junipers were planted (60 individuals for each sub-area, 20 individuals for each belt and 10 individuals for each microhabitat). We measured the survival of the juvenile junipers planted, in relation to soil salinity (3 belts from the coastline) and the microhabitat (nurse plants vs. bare soil). The second manipulative experiment was conducted in a greenhouse at the University of Sassari (Italy): the germination rate and survival of juniper seedlings based on the seed origin (2 different origins: fox dungs vs. mother plants), soil salinity (specifically, 1g NaCl/L H₂O for F1, 0.5g NaCl/L H₂O for F2, and 0.25g NaCl/L H₂O for F3) and microhabitat (2 different microhabitats: bare soil vs. dwarf shrubs – *Helichrysum italicum* subsp. *tyrrhenicum*) were examined. For each combination of seed origin × soil salinity × habitat, 3 replicates (pots) were carried out for a total of 72 pots, of which 36 were sown with seeds obtained from fox dungs collected on the field and 36 with seeds collected from adult junipers. For each salinity level, there were therefore 24 pots. Each pot was sown with 10 seeds. In the transplanting experiment, ANOVA was used to check for significant differences in the number of junipers that survived one year after planting, across two microhabitats and 3 belts from the seashore. In the second manipulative experiment, ANOVA was used to verify whether there were significant differences in the number of seeds germinated and juniper seedlings survival one year after sowing, between two seed origins, three salinity levels and two microhabitats.

In general, within the homogeneous sub-areas, an average survival of 4.2 ± 1.79 juveniles per treatment was recorded. The average survival of planted junipers was: 1 in F1, 3.75 in F2, and 2.37 in F3 in the bare soil; 4.25 (F1), 6.5 (F2), and 7 (F3) in the nurse plants. ANOVA revealed a highly significant effect of the microhabitat on the survival and growth of the junipers planted, while belt and the belt × microhabitat interaction were not significant factors. ANOVA showed a very significant effect of belt, microhabitat, and the belt × microhabitat interaction on juniper seed germination, and a significant effect related to belt, microhabitat, and the interactions seed origin × belt and seed origin × belt × microhabitat on the survival of juniper seedlings.

This study demonstrates experimentally that juvenile survival was not influenced by the salt gradient, nor by the interaction between the salt gradient and the microhabitat, but confirmed the significant role of nurse plants in enhancing the survival of juvenile trees in Mediterranean environments. Contrarily, the salt gradient and its interaction with the seed origin and the microhabitat proved to be significant factors influencing both the germination rate and seedling survival. Our findings highlight that the intricate plant-animal and plant-plant interactions are highly dependent on the specific characteristics of both the animal and plant species involved, and could be mediated by abiotic factors, such as soil salinity.

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Yeast diversity in *Onosma* spp. floral nectar and pollinators

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Floral nectar is a harsh environment, consisting mainly of water and high concentrations of sugars. Each flower functions as a distinct island habitat. Nectar-inhabiting microorganisms, such as yeasts and bacteria, alter nectar chemistry through their metabolic activities (1). Transmitted from flower to flower mainly by pollinators, these microorganisms can live under very high osmotic pressure conditions and are present in both nectar and on the mouthparts of pollinators (2, 3). However, their role in plant-pollinator interactions remains largely unexplored. This study aims to characterise yeast diversity through metagenomic analysis, isolation and identification of cultivable yeasts. Plants of the genus *Onosma* (Boraginaceae: Lithospermeae) have been used as model species, since they produce significant amount of nectar that is not diluted by rainfall due to their flower morphology. Profiling nectar chemistry using HPLC methods combined with information about the nectar microbiome could provide insight into the relationships between microbes, nectar and pollinators.

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2) C. Herrera, C. de Vega, A. Canto, M. I. Pozo. (2009). *Annals of Botany* 103 (9): 1415–23

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Landscape complexity shapes plants-arthropods interactions in agroecosystems: an eDNA approach

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The increasing simplification of agricultural landscapes, driven by intensive practices such as monoculture and habitat removal, poses a significant threat to biodiversity and the functional integrity of agroecosystems. In particular, landscape homogenization reduces ecological connectivity and impairs the capacity of agricultural systems to support essential biological processes. This issue is especially relevant in light of the recent European Union Nature Restoration Law, which establishes binding targets, including habitat restoration, reversal of pollinator decline, and the expansion of agricultural areas hosting high-diversity landscape features. In the present study, we assessed the impact of landscape homogenization on plants-arthropods interactions, through the analysis of environmental DNA (eDNA) retrieved from leaf surfaces. The study was conducted in a wine-producing region (Valle Telesina, Benevento, Italy), characterized in recent years by expansion of cultivated land. Using a systematic randomized sampling design, fifteen vineyards with similar bioclimatic conditions were selected and classified based on the proportion of their perimeter occupied by stable vegetation (hedgerows), as well as the floristic composition and structure. Twenty-five samples were collected from *Vitis vinifera* L. within the vineyards and from plant species representative of the surrounding hedgerows. DNA was extracted from leaf surfaces, amplified, and prepared for sequencing using Illumina technology. The resulting reads were analyzed using a metabarcoding pipeline based on the DADA2 protocol to identify amplicon sequence variants (ASVs). Statistical analyses were performed in the R environment. Results indicate that both the length and composition of vegetated borders significantly influence arthropod's community composition, independently of vineyard spatial distribution. The eDNA analysis detected 37 arthropod's orders (genitivo sassone vuole la 's) (including Hemiptera, Diptera, Lepidoptera, and Hymenoptera) and 554 species. We hypothesized that hedgerow structure and vegetation cover would influence arthropod's community composition, and the data support this relationship. Specifically, hedgerows with a well-developed arboreal layer more frequently hosted spiders (Araneae), whereas herbaceous and shrub layers were associated with Mesostigmata and Collembola. In vineyards lacking hedgerows, a significantly higher abundance of phytophagous mites, such as *Colomerus vitis*, and a marked decline in pollinators were observed. Conversely, vineyards with vegetated borders harbored numerous natural enemies beneficial for biological control (Acari: Phytoseiidae), suggesting a positive effect of adjacent vegetation. Our findings demonstrate that hedgerows function as refuge habitats and sources of functional biodiversity. By utilizing eDNA analysis, we showcase a powerful tool for studying plants-arthropods interactions, emphasizing the critical need to preserve diverse and structurally complex habitats to sustain vital ecosystem services in agricultural landscapes.

Palaeobiodiversity of wetlands among buried protected biotopes and glacial relicts: preliminary palynological findings and time-based indicator species from the Teor-Campomolle palaeochannel sedge peatland (NE Italy)

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The current global change is marked by an unprecedented loss of biodiversity, a major threat to ecosystems and human life. Past flora and its changes over time are increasingly vital to guide conservation efforts focused on plant biodiversity (1). Palynology is a powerful tool for studying past plant biodiversity: fossil pollen analysis can detect biodiversity shifts across centuries and millennia, along with floristic changes in relation to climate and human activity, in turn enabling to forecast future trends and thus the development of more solid ecosystem management strategies. Wetlands are among the most ecologically important areas on Earth, as initially recognized by the 1971 Convention on Wetlands. They host an incredible intersection of biodiversity, exclusive to these habitats. A long-term analysis of wetland flora is therefore essential to conservation efforts directed at them: the case study of the Teor-Campomolle meandering palaeochannel of the Tagliamento River in the distal sector of the Friuli Plain (province of Udine), which once was a sedge peatland, is exemplary in this sense. Pollen samples from a 6.5 m-long sediment core taken from the paleochannel, spanning the last 5000 years, were analysed. More than 190 pollen taxa were found throughout the series, highlighting a remarkable plant diversity, among which Cyperaceae are ubiquitous and by far the most abundant, marking this family as extremely important in this context. Three key species in this family appeared consistently and abundantly throughout the record, revealing a valuable buried biotope sealed by clay and silt deposits accumulated during the last approximately 2000 years. Traces of the EU-protected habitat type “Calcareous fens with *Cladium mariscus* and species of the *Caricion davallianae*” – now absent in the drilling area but documented elsewhere in the region – were present in the form of pollen of *Cladium mariscus* (L.) Pohl., the habitat’s flagship species, and of other taxa typical of the *Caricion davallianae* alliance, such as *Carex*, *Eriophorum*, *Juncus*, *Lythrum salicaria* L., and *Schoenus*. Pollen belonging to *Rhynchospora alba* (L.) Vahl. and *R. fusca* (L.) Vahl., glacial relict species present in the region and reportedly featured in the *Caricion davallianae* alliance in regional records (2), was also found, documenting the ancient local presence and long-term persistence of these important species. Additionally, statistical techniques, such as indicator species analysis (3), were applied to depth-based sample clusters (via CONISS), and allowed precise identification of species linked to specific single timeframes or groups of timeframes, and thus able to function as indicators of the pollen-based reconstruction of the past plant community in each of its main phases.

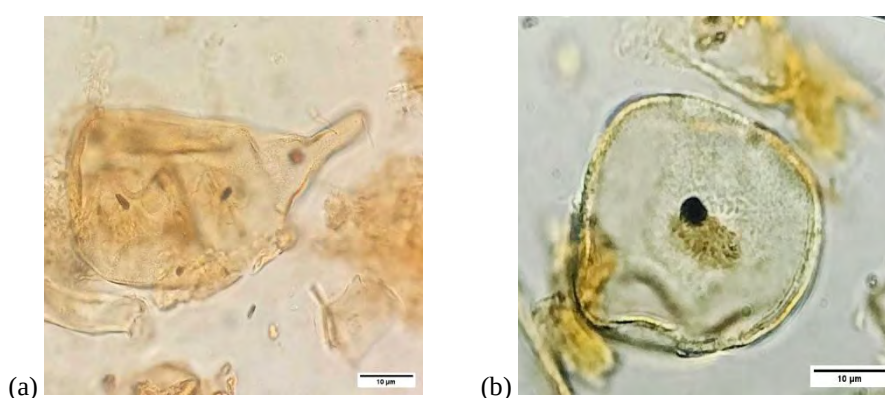


Fig. 1a-1b. Fossil pollen grains of *Cladium mariscus* (L.) Pohl. (a) and *Rhynchospora alba* (L.) Vahl. (b) from the Teor-Campomolle palaeochannel core

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The evolutionary history of the endemic palm *Chamaerops humilis* L. in the Mediterranean

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Chamaerops humilis L. is the only species of the Arecaceae family endemic to the western Mediterranean. In this study, de novo genome sequencing was used to explore possible genomic features underlying its persistence in its native range despite intense environmental fluctuations over time.

The genome was sequenced using Illumina, PacBio, and Hi-C technologies, resulting in a 3.44 Gbp assembly organized into 18 chromosomes. A total of 60,555 protein-coding genes were annotated, and repetitive elements accounted for 2.87 Gbp, nearly 84% of the total genome size. Among these, Long Terminal Repeats (LTRs) represented a major component, with the Copia and Gypsy families contributing 36.94% and 29.81%, respectively. Analysis of LTR retrotransposon insertion times revealed a wave in LTR/Copia activity around 5.5 million years ago (mya), followed by a gradual decline and a secondary minor peak approximately 2 mya. In contrast, LTR/Gypsy elements showed a steady increase in insertions from 9 to 4.5 mya, followed by a gradual decrease. These dynamics coincide with major geological and climatic events in the Mediterranean region, particularly the Messinian Salinity Crisis and the subsequent Zanclean megaflood.

Phylogenetic analyses placed *C. humilis* in close evolutionary proximity to *Phoenix dactylifera* and *Elaeis guineensis*, although only small syntenic blocks were conserved, highlighting the dynamic nature of its genome and potentially explaining the monotypic status of the genus. Gene family analyses identified several rapidly evolving families since the last common ancestor. Gene Ontology enrichment showed an overrepresentation of functions related to genome plasticity, such as transposition, cell cycle regulation, and interstrand cross-link repair, as well as stress responses, including responses to water deprivation and salinity.

Whole-genome duplication (WGD) analysis indicated a shared WGD event approximately 48.02 mya among palm species, with a later divergence from *P. dactylifera* estimated at 17 mya. The analysis of gene duplication types revealed a high proportion of dispersed duplications (41.44%) and transposon-derived duplications (28.05%), while only 13.48% of the genes originated from WGD. Several *C. humilis*-specific duplicated genes were associated with stress-response processes. The distribution of synonymous substitution rates (Ks) supported the presence of an ancient duplication event shared among the three palm species (39.87–55.2 mya) and revealed a distinct peak for transposon-derived genes, suggesting that this event contributed to the genomic architecture of *C. humilis*.

The decline in LTR insertions coincides with the end of the Messinian Salinity Crisis and the dramatic Zanclean megaflood, events that likely exerted strong selective pressures on Mediterranean flora. The expansion of stress-related genetic elements through transposition and duplication may have provided adaptive advantages during these periods of environmental upheaval.

Chamaerops humilis exemplifies a species that has endured and adapted through major paleoclimatic events, including the Messinian Salinity Crisis, the Zanclean megaflood, and successive glacial-interglacial cycles. Its genomic features likely contributed to its resilience and long-term survival in the Mediterranean basin.

The Herbarium of Brescia: Digitization, Virtual Access and Future Perspectives

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The Museum of Natural Sciences in Brescia (NE Italy) hosts an herbarium (HBBS) of ca. 70,000 specimens, of which more than 40,000 are vascular plant specimens. The herbarium includes specimens ranging from the 17th century up to date.

In order to organize and manage the specimens, the museum organized the data in a database. Each record contains information on scientific name, collector, date and place of collection, and sometimes notes about the habitat. The data were stored in a Microsoft SQL database (Bona 2012). When possible, the location of the plant was reported using the CFCE grid system (Ehrendorfer & Hamann 1965). The archive also included plant records from scientific books and articles, and from observations in the field. The database was used for writing the Flora of Eastern Lombardy (Martini et al. 2012), as well as for several other studies in the provinces of Brescia and Bergamo.

In order to improve the accessibility of the collection, the museum created an online virtual herbarium: <https://botanica.museoscienzebrescia.it>, allowing scientists, students, and plant lovers can explore the collection online. For every taxon, the system provides a dedicated page providing detailed information, such as biological form, ecological indicators, ecological strategy, legal protection (local, national, EU laws and IUCN lists), and local distribution in the province of Brescia. Furthermore, the systems host more than 30,000 high-quality images of the specimens, including about 5,500 of historical ones (e.g. Zersi, Preda, Giacomini, Arietti). One of the most important aspects of this project is that the systems allows for combining different types of data: herbarium specimens, literature, and field observations.

The whole system is based on open software (SQL and PHP) and simple web applications. This makes the entire system easy to update and manage. Furthermore, the system is easily replicable and reusable. Hence, it can be used as a model by other herbaria or museums.

Future plans include further development of the system and its interfaces, as well as tools for aggregating the data in global biodiversity platforms, such as GBIF.



Fig. 1. The Herbarium of Brescia

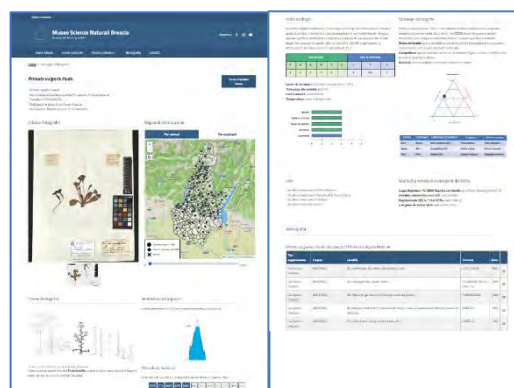


Fig. 2. Taxon sheet of *Primula vulgaris*

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Application of computer vision to the automated extraction of metadata from natural history specimen labels: A case study on herbarium specimens

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Metadata extraction from the labels of natural history collections is a pivotal task for the online publication of digitized specimens. However, it is also an extremely time-consuming task if carried out by human operators. Thus, automated (meta-)data extraction from digital images of specimens and their labels is an application where state-of-the-art computer vision techniques can be successfully applied. The task of extracting information from the labels of herbarium specimens is made of three steps: 1) text segmentation (i.e. the identification of portions of text from the image), 2) multilingual/handwriting recognition (since most of the specimens have handwritten, or at least partly handwritten labels), and 3) data parsing (i.e. the organization of the text by meaning). This study aims to explore how to transfer the general knowledge present in existing multimodal Transformers (machine learning models capable of understanding and elaborating any kind of data) into the specific sub-task of herbarium specimen label digitization. This would result in an easy-to-use, end-to-end solution, which gets rid of the bottleneck due to the limits of classic OCR (Optical Character Recognition) tools, while allowing for a high adaptability to different label formats. Donut-base, a pre-trained visual document understanding (VDU) Transformer, was the base model selected for the study. A dataset from the University of Pisa was used as a test bed. The initial attempt achieved an 85% accuracy (computed using the Tree Edit Distance - TED metric), demonstrating that the fine-tuning approach is a feasible solution, especially when compared to training the same model from scratch.

Cases in which the model had a limited accuracy were investigated to highlight possible flaws in the approach. Specimens with more than one label, especially when a mix of different handwriting and typewritten information was present, were the most difficult to deal with. Thus, possible approaches for addressing this and other small weaknesses are discussed.

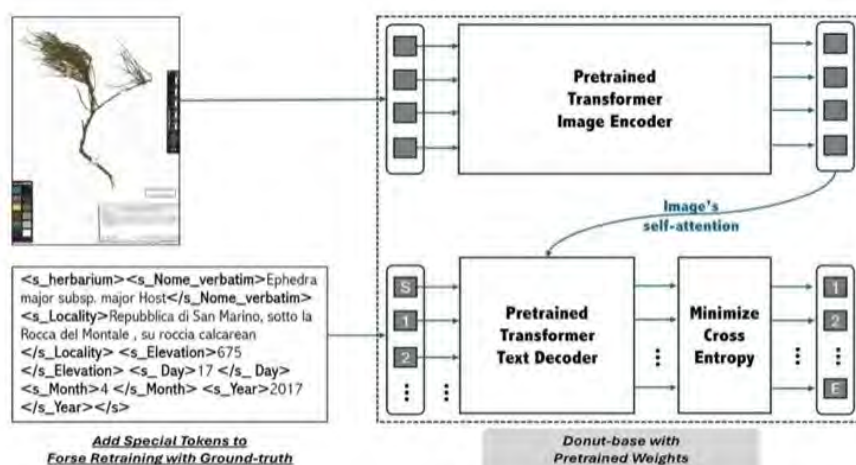


Fig. 1 Fine-tuning workflow with a set of pairs of specimen image/ground truth. Images are converted to RGB and passed as input to the pre-trained encoder for the image's self-attention. The output of the encoder is passed to the pre-trained decoder for cross-attention, matching the image with the parsed ground truth. The fine-tuning minimizes the cross-entropy loss function and forces the model to generate the same output as the ground truth

Tracking genomic shifts in alpine flora: a museomics approach to climate change assessment

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Alpine ecosystems face unprecedented warming at rates exceeding global averages, placing native plant species under extreme environmental pressure. While distributional and phenological responses to climate change are increasingly documented, genomic consequences remain poorly understood[1]. We address this knowledge gap through comparative genomic analysis of the endemic alpine species *Favratia zoysii*, using historical herbarium specimens (1780-2023) and contemporary populations. Our Illumina-based sequencing approach enables temporal comparison of genetic diversity before and after industrial-era climate shifts[2]. Our investigation aims to determine whether *F. zoysii* displays signs of positive selection in climate-responsive genomic regions, indicating adaptive evolutionary responses to environmental changes, or alternatively, exhibits decreased nucleotide diversity, and loss of rare alleles—patterns consistent with genetic bottlenecks and erosion that would suggest compromised adaptive potential in the face of accelerating climate change. To optimize species selection for comparative genomics, we developed a novel prioritization metric—the Herbarium Density Index (HDI)—that addresses a fundamental challenge in museomics research: identifying species with sufficient historical representation across relevant timeframes. The HDI combines three key parameters: temporal depth (age range of available specimens), spatial resolution (geographical distribution of collections), and sampling overlap (geographical congruence between historical herbarium sample locations and current species occurrence records, ensuring direct evolutionary comparisons across time). By integrating data from current and historic (herbarium sheets) occurrence records from GBIF, our index generates a weighted score that quantifies a species' suitability for temporal genomic comparison. This approach enabled us to efficiently screen ~3900 alpine taxa, not only identifying species with optimal historical representation for climate change genomics, but also revealing significant collection biases and taxonomic blind spots. Our research demonstrates how museum collections serve as irreplaceable genomic archives for monitoring biodiversity responses to environmental change, providing crucial evolutionary context for conservation planning in vulnerable alpine ecosystems.

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Green infrastructure as an option for *ex-situ* plant conservation

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Green infrastructure is becoming increasingly important in urban areas today. It mitigates climate change, improves the quality of life, and at the same time provides places for protecting plant species. Especially from the biodiversity point of view, it represents a green network or connection between nature on one side of urban areas and nature on the other. Botanical gardens can of course also be considered as a part of green infrastructure, whose main role is the plant conservation both *in-situ* and *ex-situ*. Especially in the *ex-situ* plant conservation, botanical gardens often encounter a lack of space, which is why we can use the innovative approach of *ex-situ* plant conservation on public green areas outside botanical gardens, i.e. in green infrastructure in urban areas. The University Botanic Gardens Ljubljana is doing the conservation of Slovenian endemic plant species on alternative sites in the city of Ljubljana, namely in the project of planting green roofs of bus shelters and on the green roofs of various institutions. As an example of effective *ex-situ* plant conservation in green infrastructure is the conservation of the endemic species *Alyssum montanum* subsp. *pluscanescens*. The species is only present in nature in one location in the eastern part of Slovenia. The population is extremely small and consists of only a few individuals. In the University Botanic Gardens Ljubljana, we successfully grew more than 200 individuals of the mentioned species from seeds and then, in a joint project with the City of Ljubljana, planted them on at least 20 bus shelters by the end of 2024. Given that the species grows on gravel in its natural habitat and is exposed to strong radiation and frequent drought, it was very suitable as a model example of *ex-situ* conservation on bus shelters. Namely, the shelters have similar environmental conditions to the natural habitat of the species *Alyssum montanum* subsp. *pluscanescens*. Based on our regular monitoring of planted species, we found that the population size of the mentioned endemic species is now larger on shelters than in nature. On each of the shelter, individual specimens are successfully spreading, flowering and also seeding. We have already observed seedlings of mother plants in the first planted shelters. This is therefore a successfully implemented *ex-situ* protection of an endemic plant species on public green areas, and there is no fear that the populations of *A. montanum* subsp. *pluscanescens* would be damaged by passers-by, as they grow on shelters that are high enough and people cannot reach them. At the same time, this is a good example of 'citizen science', as people can observe the protection of an endemic plant species from the bottom up. In a similar way, other areas in the green infrastructure of the city where there is no possibility of damaging the plant species could be used for *ex-situ* protection purposes.

Italian macroalgal herbaria in the 21st century: present uses and new roles in modern phycological research

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In Europe, marine macroalgae have been collected and preserved since the late 17th century. This practice became popular in the two subsequent centuries, when several early naturalists with interest in algae produced some of the largest algal herbaria still available today (Carl Adolf Agardh and Jacob Georg Agardh, Pierre-Louis Crouan and Hippolyte-Marie Crouan, Friedrich Traugott Kützinger, William Henry Harvey). As result, approximately one million and seven hundred thousand specimens are nowadays deposited in European herbaria (1).

In the past, herbarium specimens were mainly used for studies of algal taxonomy based on morphological features and species distributions. Nowadays they still play an important role in these research fields, in fact even more important. For instance, herbarium specimens can clarify the taxonomic identity of genera and species (2). In the last decades, the possibility to extract and sequence DNA from herbarium specimens has resulted into taxonomic revisions, discovery of cryptic species and records of non-indigenous seaweeds. Sequences obtained from type specimens have played an especially important role, as they have allowed major advancements in algal phylogeny and classification. Nowadays, however, herbarium specimens of macroalgae serve more uses than they were originally intended. Algal collections have provided information for studies of algal biochemistry, physiology and phytopathology and have become a critical resource for studies of global change biology. Furthermore, the use of herbarium data in all these scientific research fields has been greatly enhanced by the recent increasing specimen digitization and the development of online platforms.

Italian herbaria host approximately 160,000 algal specimens. Although considerably lower in number than vascular plants, these collections represent a major scientific patrimony that has proved invaluable especially for studies of historical changes in coastal marine communities. As herbarium specimens may help in detecting shifts in species ranges, new introduction, species retreat or extinction, they are indeed true witnesses of biodiversity changes.

In this framework, we present some recent research experiences based on the macroalgal herbaria's taxonomic revision and evaluation of vegetation changes. The taxonomic identity and geographical distribution of several macroalgae widespread along the Italian coasts has been clarified using sequences obtained from type specimens, even for some species that play a key role as ecosystem engineers (e.g., the coralline red alga *Lithophyllum stictiforme*; 3). For some geographical areas, such as the Lagoon of Venice, the Gulf of Trieste and the Conero Riviera, herbarium specimens allowed detailed reconstruction of historical changes in the macroalgal vegetation. For many Italian macroalgal herbaria, digitization is ongoing or planned, within the framework of several initiatives. The metadata processing and sharing through the Global Biodiversity Information Facility (GBIF), according to international standards and the FAIR principles, will further increase the value of Italian herbaria as resources for science and outreach to the general public.

1) A.M. Mannino, S.A. Minicante, C. Rodríguez-Prieto (2020) *Diversity*, 12, 309

2) D.E. Wujek, M.J. Wynne (2016) *Algae*, 31, 85-90

3) L. Pezzolesi, V. Peña, L. Le Gall, P.W. Gabrielson, S. Kaleb, J.R. Hughey, G. Rodondi, J.J. Hernandez-Kantun, A. Falace, D. Basso, C. Cerrano, F. Rindi (2019) *J. Phycol.*, 55, 473-492

Historic herbaria as cultural heritage: pre-Linnaean collections in Italian institutions

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Pre-Linnaean herbaria are essential scientific, historical, and cultural documents. The advent of Linnaeus's binomial nomenclature in *Species Plantarum* (1) marked a turning point in the classification of plants, but pre-Linnaean collections (Fig. 1) offer invaluable insight into the early stages of plant classification. In this contribution, an overview of pre-Linnaean herbaria in Italy is provided, tracing the institutions that preserve these collections, such as museums, libraries, private collections and others - while addressing their findability, curatorship and accessibility. The results give the opportunity to outline and discuss key aspects such as conservation, cataloguing, data interoperability, historical plant nomenclature, and the documentation of collection data and provenance. At the same time, opportunities for digitization are explored, especially in relation to ongoing initiatives involving high-resolution imaging, optical character recognition and the integration of herbarium specimens with archival materials. Finally, the contribution emphasizes the importance of recovering and studying these herbaria in the 21st century, advocating for a national strategy to catalogue and digitize these invaluable resources, highlighting the potential of digital tools in preserving and enhancing the value of these collections and ensuring their long-term preservation and furthering interdisciplinary research in the History of Science.

Fig. 1. One of the herbarium sheets of Andrea Cesalpino's original herbarium at the Florence University Natural History Museum botanical collections (FI), dating back to 1563

1) Linnaeus, C. (1753) *Species Plantarum, Exhibentes plantas rite cognitatas, ad genera relatas, cum differentiis specificis, nominibus trivialibus, synonymis selectis, locis natalibus, secundum systema sexuale digestas*, prima edizione, pp. 1200, + XXXI. Stockholm, Impensis Laurentii Salvii

The hidden botanical language in the ancient Roman architecture: the example of the Adrian's Arch at Jerash (Jordan)

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In antiquity, the relationship between humans and nature was related not only to the practical needs but also to the religious sphere, and any observed phenomenon was interpreted as a manifestation of divine will. This sacral dimension is evident across myths, visual culture, and material artefacts, in which plant motifs served as a real communicative language, contributing to the identity and memory of societies (1). When considering the Greek and Roman cultures, different phyto-morphic elements were used and their recurrence was adapted to the contexts of representation. The botanical identification and interpretation of the architectonical motifs is not always easy and immediate, except for few species, such as *Acanthus mollis*, *Vitis vinifera*, *Hedera helix*, *Laurus nobilis* and *Phoenix dactylifera*. Indeed, a great variety of plants, represented in the whole or through fruits, flowers, or leaves, emerges and they had a neglected significance. In this contribution, we analyse the hidden, yet well-defined presence of natural elements, that emerges through the details that form the composition of the ancient Roman architectural monuments. Here, we present, as an example, the case study of the triumphal arch of Adrian at Jerash (Jordan), which was built to honour the visit of the Emperor Adrian in 129-130 CE, enhancing the messages expressed by the numerous species represented in the capitals and in decorative mouldings (2, 3) (Fig. 1a). In the analysed motifs, we identified species like *Acanthus mollis*, *Asplenium scolopendrium*, *Lilium candidum*, *Verbascum* cfr. *thapsus*, *Asphodelus* cfr. *fistulosus*, *Nymphaea* sp., *Phoenix dactylifera* and others. Moreover, some plants were reproduced in combination, such as *Gladiolus italicus* and *Asplenium scolopendrium*, representing the hope of a military victory. Symbolically speaking, the composition of the architectural buildings represents overall concepts of Life/Fertility and the final victory of the Roman Empire, which is represented using *Romulea* sp. at the centre of a crown of *Laurus nobilis* (Fig. 1b). The evaluation of phyto-iconography and the interpretation of the hidden messages expressed through the decorations provide an innovative perspective on ancient buildings and their role in ancient societies, as well as the symbolic relevance of a naturalistic communicative language that is no longer recognised by modern society but was central in ancient ones.



Fig. 1a: on the top: botanical elements represented in the triumphal arch; 1b) on the bottom: spread representation of botanical elements and the suggested symbolic meaning

- 1) Caneva G., 2010. Il codice botanico di Augusto. The Augustus Botanical Code. Ed. Gangemi, Roma
- 2) Caneva G., Savo V., Kumbaric A., 2014 *Economic Botany*, 68(1): 109-11
- 3) Lazzara A., Kumbaric A., Pergola A., Caneva G., 2024. *Plants*, 13(19), p.2795

Occurrence of the lichen *Tephromela atra* on carbonate-rich rocks: what's so strange?

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Epilithic lichens occurring on stone artefacts are interesting subjects for the study of biodeterioration phenomena in the field of cultural heritage conservation. Lichens can seriously damage the substrate both physically, by growing hyphae between crystals, and chemically, by producing organic substances that dissolve some mineral components and eventually form new biominerals. In this study we investigated the thallus-substrate relationship in *Tephromela atra* (Huds.) Hafellner, a silicicolous lichen that occasionally occurs on carbonate-rich rocks, and identified in this case as var. *calcareae* (Jatta) Clauzade & Cl. Roux. The aim was to test the hypothesis that this colonisation only occurs when the surfaces are “superficially decalcified”, as claimed by some authors. Ten samples occurring on bioclastic limestone, calcarenite, calcareous schist and brick were selected among those preserved in the TSB herbarium. Fragments of thalli still adhering to the substrate were embedded in epoxy resin to obtain cross-sections, that were analysed by Fourier transform infrared (FTIR) spectroscopy with a Focal plane array detector (FPA) to provide chemical information at the micron scale. The sections were then stained with periodic acid-Schiff (PAS), and the percentage of hyphal coverage (PHC) was measured in selected areas of 2 mm² fixedly spaced along vertical transects from the thallus-substrate interface to the hyphal free substrate (Fig. 1). X-ray diffraction (XRD) was performed on additional lichen samples that had been pulverised in an agate mortar to detect any biomineralization products present. A selection of cross-sections was observed at scanning electron microscope (SEM). The hyphae of *T. atra* penetrated to a maximum depth of 0.8 mm in all calcareous substrates, only in brick (TSB 44405) did the penetration consistently reach a depth of 2.4 mm. The PHC varied greatly between substrates, with a minimum in compact limestone to a maximum in porous limestone. XRD analyses confirmed the presence of calcite in all samples (except TSB 44405, on brick) and showed the presence of the biominerals whewellite and weddellite in varying amount, except in TSB 44405. FTIR spectra and high-lateral-resolution chemical maps showed the presence of calcite in all specimens, with medium/high to high concentration at the thallus-substrate interface (Fig. 2). Calcite depletion was never observed. SEM observations revealed a clear continuity in the growth of *T. atra* hyphae from the medulla into the substrate layer. Here the hyphae can form dense filamentous networks between and within crystals, with direct penetration by chemical dissolution (Fig. 3). These results clearly show that *Tephromela atra* var. *calcareae* can colonize carbonate-rich rocks, not pre-decalcified.

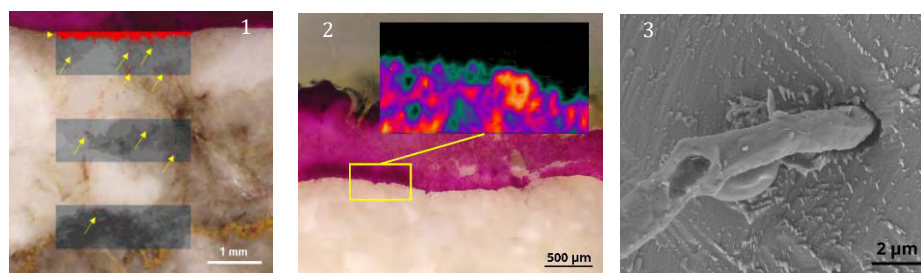


Fig. 1. Cross-section stereophotograph of *Tephromela atra* var. *calcareae* occurring on calcarenite (TSB 37937), stained with periodic acid-Schiff. The three grey areas arranged along a vertical transect were selected for the estimation of the percentage hyphal cover (PHC); the hyphae are highlighted in false colour (red) and eventually marked by yellow arrows. Fig. 2. FTIR high-lateral-resolution chemical map of calcite (TSB 35822).

Fig. 3. SEM micrograph of a hypha of *Tephromela atra* penetrating a calcite crystal by dissolution (TSB 38681, calcarenite).

Lichen recolonization on stone heritage surfaces following cleaning interventions: Should preventive strategies already consider the ongoing climate change?

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Lithobiontic communities are a recognized threat for the conservation of cultural heritage stone materials, on which they are responsible for both aesthetic decay and deterioration processes. Strategies for an effective removal of lithobionts from stone surfaces and, in particular, to prevent/slow down (re-)colonization processes after cleaning interventions are thus a major goal to improve the management of archaeological and monumental areas. However, knowledge on lithobiontic colonization dynamics and their (micro-) environmental conditional factors are still limited, particularly in the context of the ongoing climate change. With this regard, models predicted, for the end of the current century, differential trends in biomass accumulation for the northern and southern areas of Europe, with the latter affected by a significant reduction of precipitation and, consequently, a lower biomass accumulation, except for the Alps (1). However, field evidence for these patterns - which the models mostly expect for the far future at the end of the current century - still need to be recorded.

In this work, investigations were focused on lichen community dynamics in different Italian heritage sites, distributed in the Alpine, Po Plain and Mediterranean regions, and including the National Archaeological Park of Massi di Cemmo in Valcamonica (UNESCO site n. 94; Lombardia), the Walser Cemetery in Gressoney-La-Trinité (Valle d'Aosta), the Savoy Residences of Villa della Regina and Castle of Govone (UNESCO site n. 823bis; Piemonte), and the House of the Ancient Hunt in Pompeii (UNESCO site n. 829bis; Campania). In each of these sites, characterized by different climate conditions and stone substrates, lichen diversity and abundance were monitored at different intervals from cleaning interventions - which included devitalization treatments and mechanical removal of thalli- and/or in dedicated cleaned parcels. Moreover, in the case of Valcamonica and Pompeii, the photosynthetic activity of selected species was fluorimetrically monitored across different seasons, under various meteorological conditions.

In both the alpine sites in Valcamonica and Gressoney, where climate warming is testified by the recent treeline shift upwards (2), heritage surfaces showed a rapid recolonization, with lichens almost recovering the original species diversity and cover values, on the engraved rocks and decorated tombstones, respectively, within approx. 10-12 year. On the balustrades of the Savoy Residences, recolonization was primarily due to nitrophilous species characterized by abundant dispersal of spores and/or asexual propagules, while for other species, although starting immediately after the cleaning, recolonization was rather slow. In the case of Pompeii, lichen communities on differently oriented wall paintings, which did not undergo any conservative intervention, were resurveyed in 2025 after eight years, showing positive and negative trends for xero-phytic and meso-/hygro-phytic species, respectively. On S-exposed walls, in particular, a decrease of lichen colonization was observed and related to remarkably shorter periods of moisture conditions favorable to photosynthetic activity with respect to walls having a different aspect. Moreover, cleaned parcels, which for the other cardinal aspects showed evident (re-)colonization, did not show any regrowth. Such patterns seem to represent first field evidence of the expected consequences of climate change, and in particular of the reduction in days of rain, on biomass accumulation on heritage surfaces, in the southern area of Europe, although limited to the S-exposed surfaces. In analogous agreement with the predicted scenarios, lichen (re-)colonization appears particularly flourishing, rather than limited, in the Alps. Instead, patterns observed in the Po plain sites respond to different conditional factors, being likely dependent on the high eutrophication due to their urban and rural surroundings. Different urgency and strategies to address preventive interventions to contrast lichen recolonization in the different illustrated scenarios will be discussed.

1) A. Gómez-Bolea, E. Llop, E., X. Ariño, C. Saiz-Jimenez, A. Bonazza, P. Messina, C. Sabbioni (2012) *J. Cult. Herit.*, 13, 254-258

2) G. Leonelli, M. Pelfini, U. Morra di Cella, V. Garavaglia (2011) *Ambio*, 40, 264-273

The Colosseum in green – Can essential oils promote sustainable conservation?

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Cultural heritage sites are frequently affected by biofilms growth, which can lead to deterioration depending on the microbial community composition, conservation state of monument, and the climatic conditions. Controlling microbial growth is therefore essential to preserve archaeological sites from biodeterioration. This study explores the use of natural biocides, such as essential oils (EOs), as a green alternative to chemical products which are known for their hazardous nature and decreasing effectiveness due to microbial resistance (1).

To promote a sustainable conservation strategy for the Colosseum hypogeum (Parco archeologico del Colosseo, Rome, Italy), biofilm-forming microorganisms were sampled and the cultivable fractions of heterotrophs and phototrophs were isolated to perform a screening test using EOs of *Ocimum basilicum* L., *Cinnamomum verum* Presl, *Lavandula angustifolia* Mill., *Origanum vulgare* L., *Thymus vulgaris* L., and *Melaleuca alternifolia* Maiden & Betche at 5% concentration (2). Then, the EOs showing the highest biocidal activity were encapsulated in an alginate hydrogel (HG) and applied at different concentrations and exposure times to microalgal biofilms cultivated on Lecce stone. To assess the biocides effect, the photosynthetic activity was measured using a mini-PAM portable fluorometer for up to two months. The damages induced by the EOs on cell morphology were assessed by scanning electron microscopy, while the changes in cells biomolecular distribution after treatment was assessed by Fourier transform infrared spectroscopy. For preliminary *in situ* testing, Lecce stone samples were placed on the Colosseum floors to allow natural colonization by biodeteriogenic microorganisms. After characterization via metagenomic analysis, the samples were treated with selected EOs and monitored over time (Fig.1).

The results indicate that cinnamon EO was the most effective in reducing the vitality of biodeteriogens when applied in HG at 0.5% (v/v) for 48 h, leading to extensive cell lysis and affecting the biomolecular distributions. These greener approaches represent a promising alternative to traditional biocides, offering broad-spectrum antimicrobial activity and a complex chemical composition that may help to mitigate the occurrence of microbial resistance.

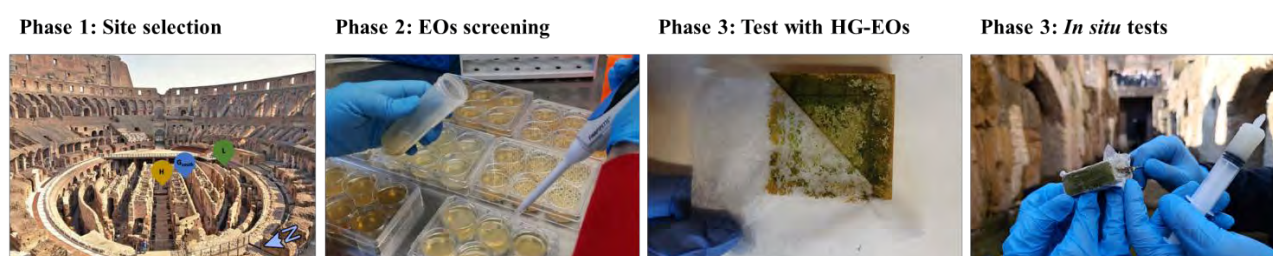


Fig. 1. Different stages of experimentation: from site selection to laboratory testing, up to *in situ* experiments. EOs: essential oils. HG: alginate hydrogel support matrix

1) P, Sanmartín., P, Bosch-Roig., D, Pangallo., L, Krakovà., M, Serrano (2023) Appl. Microbiol. Biotechnol, 107, 2027-2037

2) R. Ranaldi., L, Rugnini., G, Migliore., F, Tasso., F, Gabriele., N, Spreti., F, Scuderi., R, Braglia., P, Di Martino., A, Pujia., A, Canini (2025) Appl. Microbiol. Biotechnol, 109:48

Reconstructing coastal dune flora from 400 years ago: insights from the Aldrovandi's herbarium

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In the context of biodiversity monitoring, herbarium data provide valuable information to detect past species distributions and possible species loss. The herbarium of Aldrovandi (1551–1586), notable for its precisely documented collection locations (Baldini *et al.*, 2022), offers a unique chance to trace species distributions from XVI century, far beyond what most other sources can provide. This research aims to reconstruct the coastal dune flora from approximately 400 years ago by analysing species preserved in the Aldrovandi herbarium collection. We selected species collected from the Tyrrhenian and Adriatic coasts. We analysed species distribution, life forms, and Ecological Indicator Values for each coast. Additionally, we examined the distribution of rare species. We gathered a total of 225 herbarium samples from both coasts, corresponding to 137 species. Collection sites ranged from Albenga to Naples along the Tyrrhenian coast, with a concentration peak in the area of Pisa and Leghorn, and from Venice to Ancona along the Adriatic coast, with concentration peaks at the Venice Lido, around Ravenna, and from Pesaro to Senigallia. Exploratory analyses showed that on the Tyrrhenian coast, species with a Stenomediterranean distribution predominated (42.5%), whereas on the Adriatic coast, Eurasian (28.8%) and Eurimediterranean (25%) species were more prevalent, with Stenomediterranean species accounting for 21% only. Most of the samples collected on the Tyrrhenian coast were phanerophytes (27.5%) and therophytes (23.8%), while on the Adriatic coast most of the samples were therophytes. While we acknowledge that Aldrovandi's collecting activity may have been influenced by chance or subjective choices, we hypothesise that the more pronounced Mediterranean character of the Tyrrhenian coast may have led him to collect a higher number of Stenomediterranean phanerophyte species typical of the Mediterranean maquis (e.g., *Quercus ilex* L., *Phillyrea angustifolia* L., *Pistacia lentiscus* L.) along the Tyrrhenian coast compared to the Adriatic coast. Ecological Indicator Values for Temperature were slightly higher for the Tyrrhenian coast (mean $\pm$ SD: 8.5 ± 1.3) than for the Adriatic coast (7.6 ± 1.8), while the species indicator values for moisture showed similar values for both coasts (Tyrrhenian: 3.3 ± 2.1 ; Adriatic: 3.8 ± 2.5). The collection included endemic species, such as *Linaria purpurea* (L.) Mill. and *Limonium multifforme* Pignatti, and documented the presence of species with a currently limited distribution in Italy, such as *Thymelaea tartonraira* (L.) All. subsp. *tartonraira* (Pignatti *et al.*, 2017-2019). Our research provides a framework of the old distribution of coastal dune species and lays the foundation for further analyses on potential distribution shifts over time. In this context, the digitization of herbarium specimens is paramount for facilitating the addressing of complex questions through the use of historical data, thereby advancing research in ecology and conservation.

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Temperature-driven dynamics of *Caulerpa taxifolia* (Vahl) C.Agardh epiphytic bacterial community: implications for the alien alga diffusion in a changing climate

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In marine ecosystems, macroalgae are currently recognized as one of the main groups of invaders due to their remarkable capacity for vegetative reproduction, effective dispersal mechanisms and high adaptability to a wide range of environmental conditions, including stressful ones. These traits contribute to defining their profile as highly successful invaders (Chinchio et al., 2020). Among macroalgae, *Caulerpa taxifolia* (Vahl) C.Agardh (Caulerpaceae) is a noteworthy threatening invader in the Mediterranean Sea and it is included in the list of the worst aliens of the Basin (Streftaris & Zenetos, 2006). Although several studies on *C. taxifolia* have been conducted over time, several aspects of its complex interactions with abiotic and biotic factors remain poorly understood. In particular, data on the role of its relationships with closely interacting organisms in shaping its response to environmental conditions are still scarce in the literature (Ramanan et al., 2016). In this context, this study aimed to investigate the response of *C. taxifolia* to warming in terms of survival and health status, in relation to the qualitative-quantitative composition of its epiphytic bacterial community. To this end, two consequential experiments were performed under controlled conditions at the Civic Aquarium of Milan (Italy), during which a comparison was made for the considered aspects with the Mediterranean native congeneric *Caulerpa prolifera* (Forssk.) J.V.Lamour.

In the first experiment, the effect of temperature on thermal stress tolerance was assessed by exposing both macroalgae to three temperatures (20 °C, 24 °C and 28 °C). For each treatment thallus viability and the abundance and structure of their bacterial communities was evaluated by applying fluorescence microscopy and next-generation sequencing (NGS) techniques. The hypothesis was that a correlation exists between algal tolerance to thermal stress and the structure of its associated epiphytic bacterial community, which may play a functional role in its stress response. According to the results of the first experiment, a second experiment was conducted. *C. prolifera* and *C. taxifolia* thalli were exposed to a common intermediate temperature as well as to the most stressful one for each species, with and without antibiotic treatment. This allowed the assessment of the health status and mortality rates for algae with a significantly reduced associated bacterial community (antibiotic-treated) compared to those with an almost intact microbiota (untreated).

The results of the first experiment revealed opposite responses of *C. prolifera* (CP) and *C. taxifolia* (CT) to temperature: a higher mortality for *C. prolifera* and *C. taxifolia* (CP: 33% and CT:18%) as well as a reduction for all the traits was, indeed, observed at their suboptimal growth temperatures, of 28 °C and 20 °C respectively. Moreover, the epiphytic bacterial communities of both the species significantly changed in response to temperature, showing a greater bacterial abundance at 28 °C and 20 °C, respectively for *C. prolifera* and *C. taxifolia* (CP28: 25×10⁶ cells cm⁻¹; CT20: 30×10⁶ cells cm⁻¹). At the above mentioned suboptimal temperatures, both the algal species were associated with the bacterial genera *Suttonella* and *Bacillus*, known for their role in enhancing algal tolerance. Moreover, the second test confirmed the role of epiphytic community in algal tolerance to temperature stress because the antibiotic treatment worsened the health condition of both the species at their suboptimal temperature and especially that of *C. taxifolia* at 20 °C (CP: 75% and CT: 81%). Overall, these findings prove the role of the epiphytic microbiota in supporting the temperature adaptability of *C. taxifolia* and, more importantly, they suggest its contribution to the invasiveness of such species in the Mediterranean.

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2) Ramanan, R., Kim, B. H., Cho, D. H., Oh, H. M., & Kim, H. S. (2016). Algae–bacteria interactions: Evolution, ecology and emerging applications. *Biotechnology Advances*, 34(1), 14–29.2) S. H. Mousavi, N. Z. Tayarani, H. Parsaee (2010) *Cell. Mol. Neurobiology*, 30, 185-191

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Functional traits and plant-soil feedback mechanisms contribute to the understanding of spatial pattern formation of *Cymodocea nodosa* meadows

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Seagrasses play a vital role in coastal ecosystems by providing essential ecosystem services, including oxygen production, carbon sequestration, shoreline stabilization, and the creation of habitats for diverse marine organisms (1). At the seascape scale, in sheltered environments and in areas of recent colonization, seagrass meadows often display striking spatial patterns, such as circular vegetated patches, unvegetated gaps, or so-called *fairy circles* (2). These patterns reflect the clonal growth dynamics of seagrass populations and can undergo rapid regime shifts, including sudden collapses that lead to bare patch centers. These mechanisms can be drastically influenced by the shifting regimes of seawater intrusions as the aftermaths of sea level rise.

The emergence and configuration of self-organized spatial patterns are closely linked to the ecological condition of seagrass communities, as they respond sensitively to environmental changes. Strong plant–plant and plant–soil feedback mechanisms have been identified as key drivers of these dynamics, indicating the presence of nonlinear responses to external environmental pressures (3). However, further experimental evidence are required for identifying the drivers behind sudden pattern shifts and the consequent rapid loss of entire meadows. Disentangling these processes is crucial for assessing the resilience and vulnerability of seagrass ecosystems under changing water regime and other environmental conditions.

To investigate these dynamics, 52 meadows of *Cymodocea nodosa* (Ucria) Asch. were sampled across a vast lagoon system in the Northern Adriatic Sea. Sampling was conducted at the patch scale along a gradient of patch age, targeting the edge and the center of each patch, respectively. To characterize the age and clonal growth of the patches, patch diameter was measured and compared with historical orthophotos of the study area. At each location, above and belowground biomass, growth and functional traits (i.e. shoot density, leaf dry matter content, leaf area, leaf length, specific leaf area) of the species and sediment properties were collected. Statistical analyses were conducted using linear mixed-effects models to assess the effects of patch position and age on plant and soil variables. In addition, trait correlation networks were used to explore patterns of phenotypic integration across patch locations. Preliminary results indicated that, in the early stages of clonal growth, seagrass patches tend to expand in terms of size, increasing total biomass and shoot density. However, over the long term, a progressive loss of above-ground biomass occurs in the central location of the patches, a phenomenon that, in some cases, leads to the formation of fairy circles. In contrast, we observed a consistent maintenance of below-ground biomass in the meadows over time, regardless of patch location, highlighting the ecological resilience associated with spatial pattern modification. Finally, correlation networks among plant traits at different patch locations reveal marked changes in phenotypic integration, emphasizing the need to further investigate plant–soil feedback mechanisms through a functional approach to gain a more comprehensive understanding of the ecological dynamics driving the formation of such spatial patterns.

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Investigating habitat and functional traits of nativeness, rarity, and invasiveness in Sardinia's aquatic plants

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Mediterranean wetlands are of critical conservation importance due to their rich biodiversity, ecological functions, societal value, and the substantial declines they have experienced in recent decades (1). To support species conservation and ecosystem resilience, it is essential to understand the biological and environmental factors shaping the composition and diversity of wetland plant communities (2). This study is focused on a comprehensive assemblage of 224 vascular plant species associated with Sardinian wetlands. We analyzed 13 traits related to reproduction type, floral and seed characteristics, distribution, vulnerability and habitat preference. We used logistic regression models to evaluate which traits were explaining the nativeness, rarity, and invasiveness of such aquatic plants.

Among these, 59 of the 184 native species were classified as rare, and 21 of the 40 non-native species as invasive. Our models found that native species generally had smaller flowers compared to non-natives, which tended to flower later and reproduce more often through vegetative means. Rare native species were typically confined to narrower elevation ranges, exhibited shorter flowering periods, and showed reduced vegetative reproduction relative to more common species. Invasive species, on the other hand, were characterized by larger flowers than their non-invasive counterparts (see, for instance, Fig.1-2).

The high rates of both rarity and invasiveness observed in Sardinian wetlands likely reflect the natural fragmentation of freshwater habitats and extensive anthropogenic alterations in the region. Furthermore, variations in reproductive traits may play a key role in shaping species' rarity and invasive potential in these ecosystems. The specific focus on aquatic plant species, which often face different ecological constraints than terrestrial flora, highlights unique rarity and invasiveness dynamics. For instance, rarity in aquatic plants may be less influenced by competitive ability compared to other environments, such as mountainous areas.

Beyond offering a new checklist of Sardinian wetland flora, our study provides a valuable foundation for land-use planning and conservation actions aimed at promoting native and rare species while managing the spread of invasive ones. Notably, traits such as floral size emerged as strong but divergent predictors of both rarity and invasiveness, underlining their potential for early detection and prioritization efforts. We advocate further research into plant-pollinator interactions, particularly involving rare versus common or invasive species, as these relationships may critically influence conservation outcomes. Addressing current data gaps, especially concerning rare native species with uncertain conservation statuses, is essential. Moreover, future studies should explore additional functional traits—like seed mass and dispersal mechanisms—through direct experimental approaches, rather than inferring them from related characteristics.



Fig. 1. *Triglochin barrelieri* Loisel.



Fig. 2. *Pontederia crassipes* Mart.

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Time since first naturalization is key to explaining non-native plant invasions on islands

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Investigating the extent of insular invasions by non-native species (i.e., the number of islands they occupy) is central to island conservation. However, interrelationships among life history traits, naturalization histories, and island characteristics in determining island occupancy by non-native plants are poorly understood. We asked whether (1) island occupancy by non-native vascular plant species declines with their year of first naturalization, (2) periods of first naturalization differ among growth forms, dispersal modes, and biogeographic origins and recently naturalized species are more frequent on large, less isolated islands near urban areas. We contrasted trends across trait and biogeographic origins. We combined species lists for 767 non-native plants on 264 islands in northern Aotearoa New Zealand. We categorized species by trait and biogeographic origin and identified their time of first naturalization in Aotearoa New Zealand. Island occupancy declined with the year of first naturalization at similar rates for all categories. Herbaceous, Eurasian, and Mediterranean species, as well as those with unspecialized dispersal modes, naturalized disproportionately earlier than other categories. Non-native plants that naturalized earlier occur more frequently on large islands close to urban areas, but not on less isolated ones. Relationships with island characteristics did not differ among categories. Overall, time of first naturalization was more important than trait and biogeographic origin categories in explaining non-native plant invasion patterns on islands. Thus, management bodies should prioritize recently naturalized non-native plant categories (e.g., woody species from other regions within Oceania) and focus on large islands close to urban areas.

Urban jungle: mapping the spread of invasive alien plants across Italian cities

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With millions of people living in cities—and urban density expected to rise—understanding how urban ecosystems function is more crucial than ever, both to secure essential ecosystem services and to safeguard the biodiversity that still persists in urban areas (1). Life in the city is no easy feat for many organisms. Continuous disturbance, pollution, and the dominance of sealed surfaces over natural habitats create barriers to the establishment of numerous species. At the same time, urban contexts, with their abundance of novel microhabitats and frequent disturbances, can also act as gateways for the establishment and spread of certain species also towards natural and semi-natural habitats (2). In this regard, a mix of human-related factors and biological traits like resilience and pre-adaptation often contributes to the success of alien flora. In cities, alien plants stir debate: they are often seen as welcome greenery in concrete jungles, but, when massive, they are also perceived as a problem. In fact, some of these newcomers, thanks to human facilitation and intrinsic ecological traits, can become aggressive invaders—reshaping cityscapes in subtle and striking ways. Their impact extends to the aesthetic, ecological, and functional dimensions of urban life: invasive alien plants can damage built heritage, worsen allergies, and contribute to a sense of decay—alongside their ecological cost, such as limiting native biodiversity and hindering restoration efforts. Managing these species is increasingly relevant in urban planning, and a first step is to identify which invasive plants are most concerning, and where they establish most easily (3). To this end, we surveyed the distribution of 26 alien plant species—known for their invasiveness in Europe—across Milan, Rome, and Turin, three of Italy’s largest cities. The study focused on how specific physical properties of cities – urban elements (e.g. roads, gardens, industrial zones) and the urbanization gradient – influence their presence. Using a stratified sampling approach along an urbanization gradient, nearly 20,000 plant occurrences were recorded and analyzed. In all cities, the most frequent and abundant species resulted to be *Ailanthus altissima* (Mill.) Swingle, *Parthenocissus* spp., *Phytolacca americana* L., *Robinia pseudoacacia* L., and *Sorghum halepense* (L.) Pers., with *Ulmus pumila* L. showing unexpected dominance in Milan and Turin. Clear distribution patterns emerged: gardens and roads stood out as hotspots for alien species, whereas buildings and service areas (like industrial areas and cemeteries) were more resistant to colonization. The degree of urbanization also mattered – certain species showed a preference for highly disturbed city centres (*A. altissima*), while others were more frequent in suburban areas (*R. pseudoacacia*). This comparative analysis sheds light on the spatial ecology of invasive alien plants in Southern Europe’s densely urbanized landscapes. Our findings underscore the need for context-specific strategies that consider both plant traits and fine-scale urban characteristics. Furthermore, this work lays the groundwork for decision-support tools aimed at mitigating and preventing the spread of invasive alien plants in cities—helping urban ecosystems remain liveable, resilient, and ecologically functional. As cities continue to grow and green, understanding these dynamics is essential to strike a balance between biodiversity conservation and urban resilience.

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Counteracting the invasion of *Cenchrus setaceus* (Forssk.) Morrone for the recovery of native vegetation: an eradication project from Calabria (southern Italy)

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Biological invasions represent a major threat to global biodiversity. Invasive alien species can profoundly alter natural communities, compromising the structure and functionality of ecosystems. The expansion of these species is often facilitated by human activities and climate change, which increase the vulnerability of habitats (1). *Cenchrus setaceus* (Forssk.) Morrone (= *Pennisetum setaceum* (Forssk.) Chiov.) (Poaceae) is a perennial bunch grass native to the tropical and subtropical regions of the Old World, introduced globally for ornamental purposes and now recognized for its high invasive capacity: its current distribution includes warm-temperate and Mediterranean climate areas worldwide, including many oceanic islands. It is included among the species of EU relevance under Regulation (EU) 1143/2014, transposed in Italy by Legislative Decree 230/2017, which requires its eradication or control over the entire territory of the EU. In Italy, the species was first reported in Palermo in 1963, with subsequent expansion to Sicily and other regions, including Calabria (2).

The rapid and uncontrolled spread of *C. setaceus* is causing profound alterations in the native floristic composition, with strong competition for resources and a drastic reduction in biodiversity. The most vulnerable habitats, according to the Habitats Directive (92/43/EEC), include coastal dunes (2110, 2120), semi-natural dry grasslands (6210, 6220, 62A0) and limestone cliffs with chasmophytic vegetation (8210), often already compromised by anthropogenic disturbances such as fires, urbanization and abandonment of traditional practices. Recent studies have highlighted the strong competitiveness of this species in the xerothermic environments of Sicily and southern Italy, with similar phenomena also observed in other Mediterranean regions such as the Algarve in Portugal (3).

In this context, the research project launched in Calabria* aims to analyze in detail the invaded vegetation, comparing the results with those of other critical areas such as Mt. Pellegrino or Mt. Etna and adopting predictive models (MaxEnt) to map its current and potential distribution. The main objectives include the development of targeted eradication strategies and the definition of management guidelines, accompanied by awareness-raising initiatives aimed at stakeholders (ANAS, RFI) and citizens, to effectively counter this ecological threat.

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The role of duration and timing of induced heat waves in shaping plant community of alpine grasslands: an upscaling approach

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The Alps are experiencing an increasing frequency of summer heat waves, which threaten the stability of alpine ecosystems (1). These extreme climatic events significantly impact primary alpine grassland, which are hotspots of biodiversity and providers of crucial ecosystem services. Recent evidence proved that the combination of drought and heat stress can impact responses at species and community-level affecting the whole ecosystem response (2, 3). However, the combined effect of these extreme stresses during the growing season stages remains insufficiently understood, especially how the timing and duration of heat stress could influence the vegetation responses. This study aims to fill this gap by exploring the effects of heat waves on alpine grasslands at different growing stages assessing plant response at species and community level, using remote sensing as a tool to upscale the effect at ecosystem level.

To achieve this, we induced heat waves using shelters to completely remove rainfall while increasing temperature. The experiment was conducted on two primary alpine meadows: the first is located on the slopes of Mount Peralba (Sappada, Friuli Venezia Giulia region, Italy - 46°37'22.2"N 12°43'18.9"E - calcareous substrate), the second near the Gavia pass (Sondrio, Lombardy region, Italy - 46°20'40.1"N 10°29'51.5"E - siliceous substrate), both on a surface of about 600 m². The treatments had four levels differing in duration and timing: (1) from the beginning to the peak of the vegetative season (~ 2 months), (2) for 1 month after the resumption of the vegetation season, (3) for 1 month before the peak of vegetative season, and (4) control conditions without heat waves. Six replicates were made for each treatment. Plant responses were studied at species level, analyzing leaf traits of three major functional types: sedges, grasses, and forbs. At the community level plant biomass was investigated. We finally related plant response to multispectral vegetation (*i.e.*, NDVI, GNDVI, RGRI, ARI, CRI) indices elaborated from multispectral images collected both at close range (~ 2 m) and at UAV scales (~ 20 m).

We expect early heat waves (at the start of the growing season) to anticipate phenology and increase plant growth. Prolonged stress conditions, however, may lead to a decline in final production, triggering a premature senescence. At the species level, the most pronounced functional responses are expected in forbs under severe stress conditions. Vegetation indices derived from multispectral images collected close range are hypothesized to capture fine species and community responses, while UAV-based could be used to upscale the response at the ecosystem and landscape level.

By integrating experimental findings with remote sensing approaches, this study will advance our understanding of alpine grassland responses to climate extremes and provide critical knowledge for predicting ecosystem response, enhancing conservation strategies, and extending insights to wider alpine landscapes.

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From Folk Medicine to Functional Applications: Phenolic Composition and Bioactivity of *Stellaria media*

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Stellaria media L. (chickweed) is a widely distributed herbaceous species traditionally used in folk medicine for its soothing, anti-inflammatory, and metabolic properties. In this study, we investigated the chemical composition and in vitro biological activities of different extracts (ethyl acetate, ethanol, ethanol/water, and water) obtained from the aerial parts of *S. media*. The phytochemical profiles were characterized using UHPLC-MS/MS, which led to the identification of twelve phenolic compounds, predominantly flavonoids such as apigenin derivatives, kaempferol, tricetin, naringenin, and related glycosides. The ethanol and ethyl acetate extracts showed the highest total phenolic contents, while the ethanol/water extract was the richest in flavonoids. To evaluate the antioxidant potential, several assays were performed, including DPPH, ABTS, CUPRAC, FRAP, phosphomolybdenum, and metal chelating activity. The water extract displayed the most significant radical scavenging ability in both DPPH and ABTS assays, whereas the ethanol extract was most effective in reducing power tests (CUPRAC and FRAP). Ethyl acetate extract exhibited the highest metal chelating capacity, suggesting a diverse antioxidant mechanism across different solvent systems.

The cytotoxic activity of the extracts was tested on three cell lines (HEK 293, RAW 264.7, and HepG2) using the MTT assay at a concentration of 100 µg/mL. The water extract demonstrated the strongest cytotoxic effect on HepG2 cells (viability: 33.9%), while also significantly reducing cell viability in HEK 293 and RAW 264.7 cells. These findings highlight the relevance of *S. media* as a source of bioactive phenolic compounds with potential therapeutic interest.

Overall, the integrated phytochemical and bioactivity analysis supports the use of *S. media* as a valuable natural resource for the development of formulations in the fields of pharmaceuticals, nutraceuticals, and cosmeceuticals. Further studies could expand on its molecular mechanisms of action and explore its applicability in disease prevention strategies.

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Molecular mechanisms underlying seed germination ecophysiology of *Cardamine occulta* Hornem. (Brassicaceae) in relation to ROS production

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Seed germination is a crucial step in the establishment of a new generation, and it has been proven to be mainly regulated by the gibberellins (GAs)/abscisic acid (ABA) ratio. In recent times, another group of molecules, namely reactive oxygen species (ROS), has been identified as another major trigger of germination in dormant seeds when present in moderate concentrations. This specific range of concentrations is known as the oxidative window and is different for each species. However, the mechanistic basis for how ROS interacts with known germination-related factors (genes/enzymes) that influence the regulation of GA, ABA, and their signalling remains largely unexplored. This is also because only a few ecotypes of *Arabidopsis thaliana* exhibit dormancy and are sensitive to the effect of ROS. On the other hand, *Cardamine occulta*, an herbaceous plant native to Asia that is now spreading rapidly as an invasive species in Europe, seems to be a suitable model plant to investigate the role of ROS. According to our investigations, it exhibits a marked preference for light conditions and alternating temperatures, whereas germination is completely inhibited in the dark and/or at constant temperatures. Interestingly, under optimal germination conditions, we observed a strong accumulation of superoxide radicals at the radicle tip of the embryos. Furthermore, we found that treating seeds with methyl viologen, a ROS elicitor compound, significantly improved germination under inhibiting conditions. Conversely, the addition of ROS scavenger compounds inhibited the germination process under optimal conditions (light and alternating temperature). We also examined the effect of exogenous GAs and noticed an increase in germination for all conditions, while the addition of paclobutrazol, an inhibitor of GAs, even at low concentrations, had an inhibitory effect on germination. Interestingly, when we tested the simultaneous effect of methyl viologen and paclobutrazol, the germination was hampered, suggesting that both GAs and ROS are needed to complete the process. To deepen our investigation, we analysed the regulation of key genes involved in hormone signalling, ROS homeostasis, and the circadian clock that play a major role in germination at alternating temperatures. Our findings may extend our understanding of the physiological and molecular mechanisms involved in germination related to the production and accumulation of ROS.

Nutritional properties, chemical characterization and techno-functional analysis of *Cyperus rotundus* L. tubers

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Cyperus rotundus L. is an herbaceous monocotyledonous plant, globally recognized among the most noxious weed species as it is highly invasive, and able to avoid conventional agronomic control strategies. Its persistence is primarily attributed to a dense network of subterranean tubers, which confer adaptability to diverse environmental conditions (1). *C. rotundus* tubers are rich in a variety of bioactive phytochemicals with documented pharmacological properties. Historically, the species has been utilized for millennia as a food source (2). However, its contemporary use is largely confined to specific geographic regions, where the tubers are employed as food additives or as a dietary supplement (3). The aim of this study is to enhance our understanding of this species by characterizing the nutritional composition and techno-functional properties of its tubers, with the objective of assessing their potential applications in the food industry. Tubers of *C. rotundus* were freeze-dried and ground into a fine flour. Nutritional analysis revealed that dietary fibre was the predominant component of the flour (41.80 ± 1.31 g/100g DM), followed by starch (15.89 ± 0.63 g/100g DM), protein (9.04 ± 0.15 g/100g DM), moisture (6.58 ± 0.02 g/100g DM) and ash (4.22 ± 0.06 g/100g DM). Moreover, the tuber flour was analysed using ¹H-NMR spectroscopy, which enabled the acquisition of a comprehensive metabolomic profile. Finally, additional parameters, including pH (6.6 ± 0.01) and different techno-functional properties of the flour, were assessed to evaluate the potential applications of *C. rotundus* tubers as a food ingredient. Specifically, viscosity changes during thermal treatment revealed a peak viscosity of 614.33 ± 67.9 BE reached at 77.7 ± 2.3 °C, a breakdown viscosity of 158.0 ± 46.5 BE and an end-point viscosity of 870.33 ± 92.1 BE obtained at a final temperature of 20 °C. The flour exhibited a water holding capacity of $219.06 \pm 5.26\%$, and an oil holding capacity of $106.80 \pm 5.6\%$. Foaming capacity was $13.29 \pm 1.4\%$, with a foam stability of $64.4 \pm 3.8\%$ after 1 hour. Finally, emulsion stability analysis showed a separation rate of 2.47 ± 0.15 %/min. This study represents the first comprehensive assessment of the nutritional and techno-functional properties of *C. rotundus* tubers, aiming to support the valorisation of this plant as a novel food ingredient while proposing an alternative strategy for managing its invasiveness.

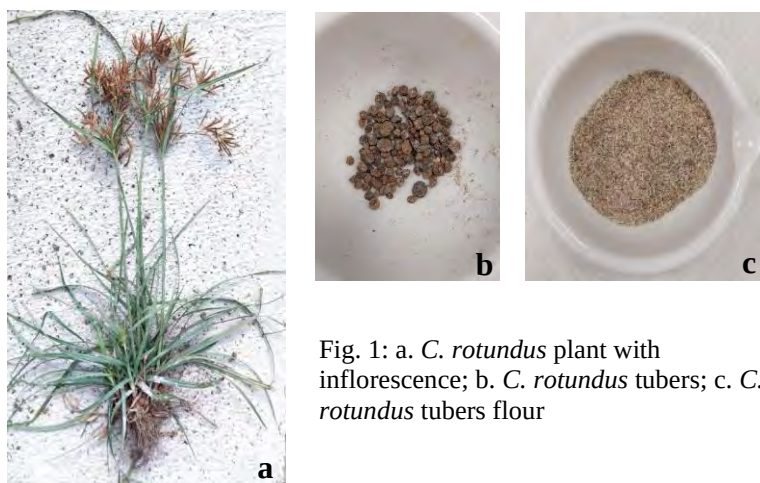


Fig. 1: a. *C. rotundus* plant with inflorescence; b. *C. rotundus* tubers; c. *C. rotundus* tubers flour

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From *Cannabis sativa* L. (Cannabaceae) by-products to bioactive agents: extraction and biological potential

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Hemp has been cultivated and utilized for thousands of years, finding applications in several industrial sectors such as textiles, nutraceuticals, pharmaceuticals, and construction. However, a gap in knowledge regarding hemp persisted for a long time, primarily due to its banishment in most parts of the world, which has been driven by its illegal use. In recent years, research on hemp cultivation and utilization has resumed, facilitated by the dissemination of non-psychoactive varieties. The PRIN 2022 PNRR project “NorCa - Not Ordinary Cannabis - P2022TXJX8 (1)” focuses on valorizing *Cannabis sativa* L. (Cannabaceae) waste and by-products from a circular economy perspective. To achieve this objective, hemp leaves were processed to extract CBD (cannabidiol) and CBG (cannabigerol) chemotypes using supercritical CO₂ (scCO₂; P: 200 bar; T: 60°C). The residual biomass was subsequently subjected to Ultrasound Assisted Extraction (UAE) with ethanol as the solvent (extraction time: 30 min, T: 60°C). The antioxidant activity of the extracts was evaluated through their DPPH radical scavenging capacity, while their antimicrobial activity was assessed against Gram-positive and Gram-negative bacteria, *Staphylococcus aureus* and *Escherichia coli*, respectively, and vs. the dermatophyte fungus *Trichophyton mentagrophytes*. Both the antibacterial and antifungal activities were evaluated following the CLSI (Clinical and Laboratory Standards Institute) standard microdilutions method to determine the Minimal Inhibitory Concentration (MIC) (2). The results pointed out that the antimicrobial efficacy depended on the extraction method used. Specifically, CBG and CBD chemotypes extracted by scCO₂ showed significantly lower MIC and IC₅₀ values than those obtained through UAE approach. For instance, against *S. aureus*, the scCO₂ extracts based on CBG and CBD as the main compounds exhibited MICs of 50 µg/mL and 100 µg/mL, respectively, with IC₅₀ values of 10.102 µg/mL and 27.045 µg/mL. Conversely, the UAE extracts demonstrated a MIC of 500 µg/mL and an IC₅₀ of 128.24 µg/mL for CBG, whereas CBD extract achieved a maximum growth inhibition of only 42.41%. A similar trend was observed for *E. coli*, although the MIC and IC₅₀ values were higher than those for *S. aureus*. Along with the standard inhibition of *T. mentagrophytes* conidia germination via microplate assay (microdilution method) (2), the antifungal activity was further evaluated following the EUCAST (European Committee on Antimicrobial Susceptibility Testing) standard diametral growth inhibition method (3). The diametral growth was observed after 7 days, 14 days as well as at the negative control confluence. The maximum growth inhibition was recorded at 14 days at the highest concentration of 500 µg/mL, with an inhibition of 60.25 ± 1.36%. In addition, after 6 days of incubation, MIC values were determined using the microdilution method, obtaining MIC values of 250 µg/mL and 125 µg/mL for CBD and CBG extracted by scCO₂, respectively, while no inhibition was observed for UAE extracts. The antioxidant activity showed a higher efficacy in extracts obtained through UAE compared to those obtained through SFE. Particularly, the IC₅₀ values were 539.13 ± 46.37 µg/mL and 533.17 ± 52.17 µg/mL for CBG and CBD extracted by SFE, respectively, while the UAE extracts yielded IC₅₀ values of 200.97 ± 11.19 µg/mL and 204.14 ± 12.21 µg/mL for CBG and CBD, respectively. In conclusion, there is almost no difference between the two chemotypes and the extraction methods regarding the antioxidant activity of the extracts. Conversely, the antimicrobial activity exhibits a significant variation depending on the cannabis chemotype and on the extraction approach performed on the plant material.

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Antioxidant properties of three *Lavandula* genus essential oils compared to a commercial essential oil

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The genus *Lavandula* L. (lavender) consists of 39 species, including *L. angustifolia*, *L. grosso* and *L. super* [1-2]. Lavender essential oils (EOs) are known for their many applications and properties. The project aimed to compare the phytochemical profile and the antioxidant activity of three different lavender EOs (*L. angustifolia*, *L. grosso*, *L. super*) in comparison to a commercially available lavender essential oil. The phytochemical profile was determined by GC-MS. The EOs cytocompatibility and antioxidant potential were determined by cell viability tests on a human neuroblastoma cell line (SH-SY5Y) in the presence or absence of H₂O₂, used as an oxidative stimulus. Subsequently, the expression levels of two genes involved in oxidative protection, NQO1 and CYT-B, were evaluated after a 12 h exposure to *L. grosso* and *L. super* EO at 0.0001%. Interestingly, the EOs had a comparable chemical profile, mainly containing linalool and linalyl acetate. However, traces of chemicals used as fragrance additives were found in the commercial essential oil. were found to be cytocompatible. Furthermore, the treatment with *L. vera*, *L. grosso* and *L. super* EOs significantly increased cell viability in the presence of H₂O₂, whereas the commercial lavender EO did not show the same effect. The antioxidant potential of the two more promising lavender EOs was better investigated; in particular, the treatment with *L. grosso* and *L. super* EOs significantly increased the expression of NQO1 and CYT B, with *L. super* outperforming *L. grosso*. In conclusion, this study confirmed the antioxidant activity of lavender essential oils, highlighting also the central role of the quality evaluation of commercial EOs, as added adulterants could affect their biological activity.

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Analysis of sugars, amino acids and phenolic compounds in nectar of selected melliferous native species in Slovenia

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Plants provide reward for pollinators in the form of nectar. Sugars, amino acids and phenolic compounds are among the most represented substances in nectar. Glucose, fructose and sucrose are three main sugars and based on the sugars ratio and nectar quality in general we can determine which pollinator prefers which flowers. We investigated nectar of 12 plant species that are native in Slovenia. We chose three plant families (Rosaceae, Lamiaceae, and Ranunculaceae) that are known as more melliferous. Plant species were chosen based on their flowering, because we wanted to research nectar through all year. Our researched species are: *Helleborus niger*, *Anemone nemorosa*, *Caltha palustris*, *Fragaria vesca*, *Ajuga reptans*, *Crataegus monogyna*, *Prunus spinosa*, *Salvia pratensis*, *Filipendula vulgaris*, *Prunella vulgaris*, *Salvia glutinosa* and *Clematis vitalba*. We also investigated how abiotic factors (air and soil humidity, air and soil temperature and UVB – radiation) and time of the day effect on quantity and quality of nectar of each plant species.

The field research was conducted from 2023-2024 in meadows and the edges of the forest around Ljubljana, Slovenia. Lab research (analysis of sugars, amino acids and phenolic compounds) was conducted from 2023-2025. We obtained the nectar with two different methods – sucking nectar with microcapillaries and method with washing flowers. Through our research we observed the advantages and disadvantages of both methods. Our research showed that nectar quantity is not conditioned by plant families or flower size, but mostly with time of the day and some abiotic factors. As for the quality of nectar we find that is mostly conditioned by families. We analysed sugars with a UHPLC system coupled with a charged-aerosol detector (CAD), phenolic acids with UHPLC system coupled with a UV detector and amino acids with HPLC system coupled with a TSQ Quantum mass spectrometer with an electrospray ion source.

With the knowledge about nectar composition we can help to plan bee gardens for beekeepers and to raise awareness about the importance of colourful meadows.

Reproductive mechanisms in *Ginkgo biloba*: male cone development, pollen formation, and the intricate crosstalk between male and female gametophytes

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The reproductive biology of gymnosperms remains one of the least understood chapters in seed plant evolution, despite their critical phylogenetic diversity and position. Among them, *Ginkgo biloba*, the only extant species of Ginkgophytes, can offer important insights into the reproductive strategies of spermatophytes. Our research leverages this unique position to investigate the molecular and developmental mechanisms underpinning male cone development, pollen formation, and the crosstalk between male and female gametophytes.

Ginkgo reproduction unfolds over several months, with a notable temporal separation between pollination and fertilization. During this long period, the male gametophyte continues its development inside the ovule, nourished via a specialized haustorial pollen tube that penetrates the nucellar tissue. We explore this process through detailed analyses of pollen formation, cellular ultrastructure, hydration-dependent morphological changes, and germination inside the ovule.

To uncover the molecular signals mediating the prolonged interaction between the male and female gametophytes, we hypothesize a complex multi-step recognition system involving surface molecules on the pollen exine, exposed intine components such as arabinogalactan-proteins, and *de novo* synthesized effectors. By conducting comparative chemical profiling of *Ginkgo* pollen and that of other gymnosperms, we aim to place *Ginkgo*'s unique features within a broader evolutionary framework of seed plant reproduction.

Our investigation encompasses male reproductive structures development, identifying distinct stages in cone morphogenesis: dormant winter buds containing male cones primordia, dividing microspores stage during bud opening, and pre-dehiscent mature cones. Hormone localization assays suggest that auxin may play a pivotal regulatory role during cone morphogenesis, a finding further supported by transcriptomic analyses that are beginning to unravel the gene networks orchestrating these developmental transitions. In particular, we are focusing on a comparison between *Ginkgo* and their close relatives among Cycadales to identify conserved and divergent mechanisms underpinning male gametophyte formation and cone morphogenesis.

By integrating structural, molecular, and physiological approaches, our work provides a comprehensive view of male gametophyte development and its coordination with ovule responses in *Ginkgo biloba*, leading to a dynamic model in which pollen-derived signals actively influence ovule development and reproductive success.

Brassinosteroids seed-priming to enhance *Sorghum bicolor* salt tolerance

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Crop productivity and ecosystem survival is constantly and increasingly threatened by the soil salinization phenomenon (Atta et al., 2023). Climate change is reducing traditional European cereal crops productivity year by year, *Sorghum bicolor* is a salt-tolerant crop mainly cultivated in arid and semi-arid regions of the world and could be an interesting alternative for European agriculture. Despite its natural resistance to osmotic stress, there are differences in salt-tolerance among genotypes (Peduzzi et al., 2024) so it is necessary to find a way to help the more sensitive genotypes to cooperate with salt stress without harming the environment. Brassinosteroids (BRs) are known to be involved in plant responses to many stresses, including salt stress. These steroidal hormones could be exogenously applied to enhance crops resistance using many techniques and one of the most promising is the seed-priming technique (Neha et al., 2022). Seed-priming provides a double advantage to the crop, both economic and ecological: the direct application on the seed allows the use of a reduced quantity of the compound and avoids its spreading into the environment. In this study, we tested the efficacy of seed priming with 24-epibrassinolide, a bioactive BR precursor, to reduce the negative effects of 150 mM of NaCl in two sorghum hybrids, a tolerant and a sensitive one, namely Bianca and Tonkawa. The seedlings were analysed at two different time points: 6 days and 10 days after sowing. We studied the germination rate of the seeds, the morphology of the root system, the adventitious roots anatomy, the gene expression for the main vacuolar sodium transporter (*SbNHX2*) both in root and shoot with RT-qPCR and the sodium inside the root cells was localized with confocal microscopy. The germination rate was significantly increased by the priming for the Tonkawa genotype in presence of salt. The morphological analyses revealed that the priming implemented the number of adventitious roots and lateral roots primordia in both genotypes. The organization of the root tissues of the sensitive genotype under salt stress was restored by the priming treatment. The relative expression of the main sodium transporter was enhanced in both genotypes by the priming treatment, in presence of salt stress. The localization of sodium in the root cells confirmed that there was a difference between the two genotypes in the ability of sequester Na^+ in the vacuole and that the priming helped the sensitive one deal with this task.

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Resurrection Plants and their Volatilome: the unresolved mystery of BVOC emissions under extreme desiccation

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Resurrection plants possess an extraordinary survival strategy against desiccation, enabling them to withstand extreme dehydration of their vegetative tissues (Farrant et al., 2007). They can lose up to 95% of their water content and, upon rehydration, resume normal physiological functions. This makes them ideal model organisms for studying how crops might be developed to survive drought conditions. Among their suite of biologically active molecules, some of these phytochemicals may also be volatile. Despite extensive studies on their metabolic and physiological adaptations, the emission of biogenic volatile organic compounds (BVOCs) from these species has been poorly investigated, especially in relation to their survival strategy during extreme physiological desiccation and subsequent recovery after rehydration (Rapparini et al., 2024).

In this study, the leaf emission of BVOCs was investigated in the resurrection angiosperm *Haberlea rhodopensis* Friv. (Gesneriaceae), a homoiochlorophyllous species characterized by the retention of the photosynthetic apparatus and pigments, which enables a faster recovery upon rehydration. However, the high concentration of chlorophyll during desiccation can lead to the production of harmful reactive oxygen species. Therefore, *H. rhodopensis* requires enhanced antioxidant protection against free radical attacks experienced either during dehydration or immediately after rehydration. Our study aims to characterize the volatilome of *H. rhodopensis* and elucidate its potential role in survival strategy of this resurrection species. Volatile emissions from the leaves of *H. rhodopensis* during dehydration and rehydration were monitored using different dynamic headspace systems, in combination with both offline chemical analysis of volatiles via gas chromatography–mass spectrometry (GC-MS) and online temporal analysis of BVOC emissions using proton transfer reaction time-of-flight mass spectrometry (PTR-MS). Isoprene emission was detected and found to vary throughout the dehydration/rehydration cycle. Isoprene may play a role in the early stages of rapid desiccation, when photosynthesis is still active and drives carbon to isoprene synthesis. The relevance of other BVOCs -such as volatiles from the lipoxygenase (LOX) pathway (e.g., C6 green leaf volatiles) and short-chain oxygenated compounds (oxVOCs, e.g., acetone and methanol)- was also highlighted. The detection and identification of novel compounds, i.e. short-chain apocarotenoids, in the volatilome appear relevant as these volatiles have recently been recognized as new signaling molecules enhancing plant tolerance to drought (Havaux et al., 2020). Our investigation into the volatilome of *H. rhodopensis*, alongside concurrent analysis of photosynthetic activity and photoprotective mechanisms, provides new insights into the potential roles of BVOCs for the “drying without dying” ability of resurrection plants.



Fig. 1. *Haberlea rhodopensis*

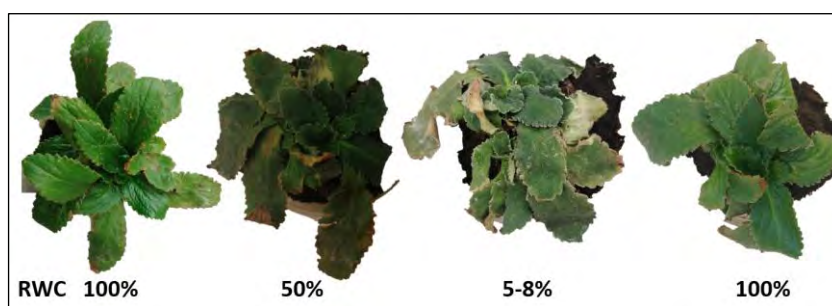


Fig. 2. Dehydration/Rehydration cycle of *H. rhodopensis*

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Phytotoxicity of *Zingiber officinale* Roscoe aerial biomass: Bioactive potential for sustainable weed suppression

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Zingiber officinale Roscoe (Zingiberaceae), commonly known as ginger, is widely used as a spice and medicinal plant. While the rhizomes are the main commercial product, the aerial parts of the plant are typically discarded after harvest. In the framework of a circular economy, these residues represent a promising source of bioactive compounds with potential applications in sustainable agriculture. This study investigates the allelopathic potential of ginger aerial biomass for the development of natural herbicides aimed at reducing reliance on synthetic agrochemicals.

Aqueous extracts and powdered material derived from ginger leaves were tested on four weed species (*Echinochloa oryzoides* (Ard.) Fritsch, *Lolium multiflorum* Lam., *Sinapis alba* L., *Trifolium incarnatum* L.) through pre- and post-emergence bioassays under controlled conditions. The treatments were applied on both filter paper and soil substrates to simulate different environmental contexts. Pre-emergence trials revealed strong phytotoxic effects, with complete inhibition of seed germination observed in *L. multiflorum*, *S. alba*, and *T. incarnatum* for both powder and extract at the highest concentrations. Although *E. oryzoides* showed partial tolerance, its germination and seedling development were significantly compromised. Post-emergence foliar application of the aqueous extract caused visible symptoms of toxicity in all species tested, especially in *E. oryzoides* and *T. incarnatum*, confirming the potential of ginger residues for direct use in weed suppression strategies.

Chemical profiling of the aqueous extract by UPLC-HR-MS revealed a complex mixture of flavonoids and phenolic compounds, including vicenin-2 and kaempferol-3-O-rutinoside. SPME-GC/MS analysis of volatile fractions showed sesquiterpene dominance, with β -caryophyllene as the most abundant component. These results highlight the allelopathic potential of ginger residues and their application as bioherbicides in organic and low-input farming systems, contributing to the circular use of agricultural by-products and the reduction of synthetic herbicides.

Differentiation of seed germination traits along a small-scale hydroperiod zonation in Mediterranean temporary ponds

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Understanding the regeneration strategies of ephemeral vegetation in Mediterranean temporary ponds (MTPs) is crucial to achieve a deeper view on the possible effects of shifting water regimes on this endangered habitat, whose conservation is considered a priority in Europe (Council Directive 92/43/CEE; code 3170*). In this study, we tested the hypothesis that germination traits vary along the small-scale plant zonation that characterizes MTPs. Despite being a highly variable habitat, three different zones related to the hydroperiod can indeed be distinguished in MTPs: a long-lasting flooded area, followed by a short-lasting flooded area, and finally the pond's outer belt (Fig. 1).

Mature fruits or seeds (hereafter, 'seeds') of 13 species were collected along the zonation of MTPs in the Hyblean plateau (south-east Sicily, Italy). The germination response to different levels of temperature, thermoperiod (constant and alternating), light, and after-ripening was evaluated for each species. Using a thermal-time approach, base (T_b), optimal (T_o), and ceiling (T_c) cardinal temperatures for germination were calculated. The effect of each factor on final germination was analyzed using MCMCglmm models, accounting for the role of phylogeny, seed mass, and pond zonation. The shape of the germination niche across zones was evaluated with ordination analysis (PCA), performed on the species×seed traits matrix.

Temperature preference varies significantly along the zonation, with species from the outer belt exhibiting higher T_o and T_c values than those from flooded areas. Furthermore, species from the long-lasting flooded area showed the highest germination percentages in darkness. Alternating temperatures and after-ripening, on the other hand, had a generally positive effect across all zones. At the species level, the ordination analysis pointed out a high variability in light and thermoperiod preferences among species from the short-lasting flooded area. Our findings support the hypothesis that temperature and light preferences for germination vary across the small-scale plant zonation of MTPs. Under current climatic conditions, this allows species from the outer belt to be among the first to germinate, while species from long-lasting flooded areas can germinate even under turbid water. The different thermoperiod and light preferences observed in species from the short-lasting flooded areas may reflect adaptations to cope with more pronounced fluctuations in water regimes characteristic of this intermediate zone. With projections of increased out-of-season rainfall and altered precipitation seasonality in the Mediterranean, our results highlight the importance of using germination traits to better understand the ecological dynamics of wetland plant communities under climate change.

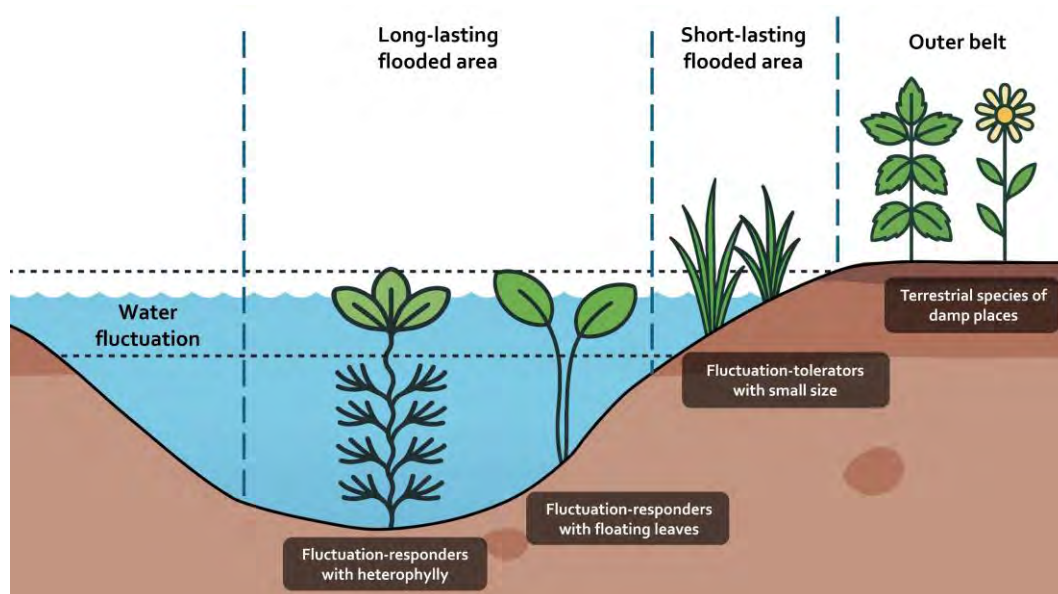


Fig. 1. MTPs' small-scale plant zonation and functional groups of plants investigated

Wood distillate as a biostimulant for mitigating drought stress in basil: effects on growth, physiology, and nutrient balance

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Water stress is one of the most critical abiotic factors limiting plant growth and agricultural productivity worldwide. As climate change intensifies, periods of drought are becoming more frequent and severe, posing a serious threat to food security and sustainable farming. Under water-limited conditions, plants suffer from reduced photosynthesis, impaired nutrient uptake, oxidative damage, and growth inhibition (1). To mitigate these effects, there is increasing interest in the use of biostimulants, natural substances or microorganisms that enhance plant resilience and performance under stress. Biostimulants can improve water use efficiency, stimulate antioxidant defenses, and support physiological processes essential for plant survival and productivity. Among these, wood distillate (WD), a by-product of biomass pyrolysis rich in organic acids and phenolic compounds, has shown promising potential in alleviating drought-induced stress in crops (2).

This study investigated whether the foliar application of wood distillate (WD) affects key parameters, such as fresh weight, photosynthetic efficiency, antioxidant compounds, stress-related biochemical markers, and mineral content in basil plants, used here as a model crop grown under water-limited conditions. The experimental design included a control and WD treatments (applied via foliar application at 0.2%) under three drought levels: no stress, moderate stress, and high stress. The results indicated that the application of WD improved fresh weight and chlorophyll content, reduced oxidative stress, and helped maintain stable levels of essential nutrients across varying drought intensities. These outcomes highlight the potential of WD as an effective biostimulant for enhancing drought tolerance in basil plants under water-deficient conditions.

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Comparison of morphological and molecular analysis method for truffle plants used in Spain, France and Italy

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Truffle farming is an activity that is gaining increasing interest to make up for the decline in natural truffle production and as a nontraditional and sustainable agriculture. Truffle cultivation mostly depends on high quality standards of mycorrhizal truffle plants and yet must then be planted in soil with suitable environment and cared for properly. Therefore, it is necessary for truffle plants to undergo serious inspection by third-party bodies to certify their suitability for truffle cultivation (Fig 1). The objective of the research was to evaluate the effectiveness and reliability of the most widely used certification methods and to provide a comparative evaluation of the performance of each method. In this research, an analysis of plants from two nurseries, one Italian and one Spanish, was conducted using two host-plant combinations, *Quercus ilex* L. inoculated with *Tuber melanosporum* Vittad. and *Corylus avellana* L. inoculated with *T. aestivum* Vittad.. These two plant species were chosen because they have very different root development, allowing to assess whether the methods work effectively under both conditions.

Three different evaluation methods (1) Fischer and Colinas, (2) Donnini et al., (3) Chevalier and Grente were applied to each of the 48 plants, 12 truffle-seedlings for each of the two plant-truffle combinations from both nurseries.

Subsequently, the results were statistically analyzed in order to determine whether there was a significant difference in the ratings obtained from the different methods. The results of the research showed that although the methods seemed to agree in the evaluation of each with high or low levels of root colonization, there were significant differences between the different evaluation methods. For example, (3) Chevalier and Grente's method was found to be more influenced by the experience and skills of the analyst, while the methods proposed by (2) Donnini et al. and (1) Fischer and Colinas proved to be less subjective and more reproducible. In addition, the Italian protocol emerged as the most effective method in detecting contaminants, able to detect them even at low concentrations not detected by the other two methods.



Fig. 1. Mycorrhizal root system

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Effect of Heat, Drought, and their interplay on Stem and Root Cambium cell activity in *Populus nigra* L.

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Climate change is reshaping plant growth dynamics, prompting numerous studies on the responses of forests and plants to these changes. Global temperatures have already risen by 1.1 °C compared to pre-industrial levels and are projected to exceed 1.5 °C within the 21st century. In particular, Extreme Weather Events (EWEs), such as heatwaves and droughts, significantly affect plants. Unlike climate change, EWEs are sudden, localized events that highlight the need to understand their ecological impacts (1). Indeed, when extreme heat and prolonged drought occur together, the stress on plants intensifies, and studying these events separately may underestimate their combined impact on plants.

The vascular cambium (VC), a lateral meristem, drives radial growth by producing xylem inward and phloem outward, which are essential for transport and mechanical functions. In temperate forests, this activity is seasonal, starting in spring when large vessels form for water and nutrient transport (earlywood) and ending in autumn (latewood) as small cells with thick walls develop before ceasing completely in winter (2). The activity and differentiation of the VC are regulated by both internal (e.g. hormones) and external (e.g., temperature, water) factors and may also exhibit different patterns above and below ground due to the soil buffer effect (3). During the growing season, environmental stressors such as drought or elevated temperature may cause VC activity to cease temporarily, leading to the formation of false rings or intra-annual density fluctuations (IADFs) in the xylem tissue. Upon the return of favorable conditions, growth may resume, although prolonged stress can lead to plant death (2).

This study aimed to assess how VC of *Populus nigra* L. responds to the combined effects of heat and varying levels of drought. We hypothesized that (i) VC activity and recovery decrease in magnitude with increasing drought severity, (ii) the combined effect of heat and drought has a stronger inhibitory impact on VC activity during the stress period and on VC recovery after stress release, compared to the effect of each stressor alone, (iii) the root and stem VC exhibit different activity patterns under heat and drought conditions.

To test this multiple hypothesis, we conducted an experiment on two-year-old *Populus nigra* under drought, heat, and combined stress. Plants were placed in two different growth chambers, one at optimal temperature (20–25 °C) and the other at higher temperature (35–30 °C). In each chamber, three cohorts of well-watered plants were progressively exposed to moderate, severe, and extreme drought, while one cohort was kept well-watered. After reaching each drought level, a subset of plants was rewatered until field capacity and measured after 10 days. Two centimeters of stem tissue above and below the collar were collected, embedded in parafilm, sectioned, and stained. Afterward, the thickness of the cambial and derivative cells, as well as the non-collapsed phloem, were measured.

Preliminary results suggest that extreme drought alone dramatically reduced the VC characteristics. When combined with heat, both severe and extreme drought levels caused irreversible damage to VC function in both the stem and root. However, the root VC appeared slightly less affected by these stressors on VC. Plants under moderate and severe stress, once rewatered, showed high levels of VC activity comparable to those of control plants. Moreover, neither heat nor drought alone resulted in permanent effects on VC activity.

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Precipitation-driven variability in VOC and SVOC emissions in the carnivorous plant *Drosera rotundifolia* L. (Droseraceae)

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Drosera rotundifolia L. (Droseraceae) is a carnivorous plant with a broad Holarctic distribution, typically occupying nutrient-poor, acidic peatlands (1). To survive in these challenging environments, the species has evolved an array of morphological and chemical adaptations facilitating insect capture and digestion. While the structural mechanisms of carnivory in *D. rotundifolia* are well-characterized, the ecological role of volatile organic compounds (VOCs) and semi-volatile organic compounds (SVOCs) in prey attraction remains insufficiently understood, particularly in relation to environmental variability (2). In this study, we investigated how climatic factors, especially precipitation and humidity, influence VOC and SVOC emissions and prey capture efficiency in *D. rotundifolia*. We collected 39 individuals from seven natural populations across two climatically distinct regions of northwestern Spain: Serra do Cando (characterized by a wet Atlantic climate) and Sierra de Ancares (with a drier, Mediterranean-influenced climate). Following transplantation to a greenhouse common garden, all plants were maintained under standardized conditions and analyzed for prey capture and chemical emissions. Prey capture was quantified via stereomicroscopy. VOCs were extracted via solid-phase microextraction (SPME) and analyzed using gas chromatography-mass spectrometry (GC-MS). SVOCs were obtained by hexane rinsing of leaf surfaces and similarly analyzed by GC-MS. Our results reveal significant inter-population differences in both the abundance and composition of emitted compounds. Notably, plants from the drier region produced more concentrated VOC profiles, potentially enhancing their ability to attract insect prey under less favorable conditions. Emissions were dominated by sesquiterpenes and sesquiterpenoids, indicating a conserved yet environmentally responsive chemical signature across populations. Despite these chemical differences, prey capture rates did not significantly differ between the two regions. One possible interpretation is that increased VOC emissions in drier environments serve to offset reduced insect availability, thereby allowing plants to maintain equivalent foraging success. In contrast, SVOC profiles exhibited less pronounced variation among populations, suggesting a more stable functional role, possibly related to short-range signaling or surface-level interactions. These findings support the hypothesis that VOC emissions in carnivorous plants are plastic traits shaped by local climatic pressures. However, their functional impact on prey acquisition appears contingent upon broader ecological contexts. This study contributes to a growing body of evidence emphasizing the interplay between abiotic factors and plant-insect interactions, and underscores the potential of VOCs as adaptable tools for nutrient acquisition in carnivorous species (3).

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Integrating Seasonal Translocation Strategies for the Conservation of *Lobaria pulmonaria* in Mediterranean Woodlands: Insights from the BioConLobaria Project

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Effective lichen conservation requires a multifaceted approach that considers the complex interplay between species-specific biological traits, habitat characteristics, site selection, interactions with other organisms, and environmental variables such as microclimate, substrate conditions, and pollution levels. Among macrolichens, *Lobaria pulmonaria* (L.) Hoffm., commonly referred to as the “lung lichen,” represents a model species due to its ecological sensitivity and its established use as a bioindicator of forest habitat continuity and quality. Despite being widely studied in boreal and temperate regions, *L. pulmonaria* remains poorly investigated in Mediterranean environments, where habitat fragmentation, intensive forest management, and climate change pose significant challenges to its persistence. Given its vulnerability, *L. pulmonaria* is of particular conservation interest, and its successful translocation could offer broader benefits for the preservation of other vulnerable species.

The BioConLobaria project addresses these challenges through a comprehensive programme based on seasonal translocations of *L. pulmonaria* across Italy: Emilia-Romagna (northern Apennines), Tuscany (central pre-Apennines), and Campania (southern Apennines). The project is structured around four core objectives:

- 1) to determine the influence of seasonal timing on the success and resilience of translocated thalli.
- 2) to evaluate the role of population stage (young vs. adult thallus fragments) in translocation outcomes.
- 3) to explore the impact of genetic diversity on the viability and acclimation of translocated individuals.
- 4) to assess receptor site suitability, identifying microhabitat characteristics that promote long-term survival and establishment.

The experimental design simulates conservation-oriented translocations by relocating fragments of both young and adult thallus fragments from source populations into two contrasting types of forest habitats: (i) mature, ecologically stable stands of oak (*Quercus* spp.), chestnut (*Castanea sativa*), or beech (*Fagus sylvatica*), and (ii) disturbed environments, such as selectively logged stands represented by isolated trees of the same forest type.

Translocation events are performed in different seasons to identify the most favourable period for ensuring the survival and health status of the transplanted thalli. Each translocated sample is subjected to one of three treatments:

- 1) short-term seasonal acclimation (3 months),
- 2) medium-term exposure (1 year), and
- 3) permanent translocation for future monitoring beyond the current project scope.

To monitor translocation success, we assess a range of morpho-physiological and structural parameters, including thallus surface area, hydration status, water-holding capacity, chlorophyll content, and photosynthetic efficiency. These are measured non-destructively both pre- and post-translocation, allowing for an evaluation of physiological acclimation and vitality. In parallel, microclimatic conditions (temperature, relative humidity, light availability) and thallus hydration are continuously recorded using data loggers at each receptor site.

By March 2025, the initial seasonal cycle of the project had been completed, concluding the first year of trimestral sampling. The preliminary results reveal significant differences in growth rates and physiological performance depending on the season of translocation, population stage, forest type and grazing effects by terrestrial gastropods. These early findings demonstrate that translocation success is not only contingent on abiotic conditions but also on biotic interactions and the developmental stage of the transplanted material. By adopting a long-term, experimental approach that integrates ecological monitoring with physiological assessment, BioConLobaria aims to establish evidence-based guidelines for future lichen conservation interventions in Mediterranean forests and beyond.

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ddRAD sequencing: a valuable method for assessing genetic diversity in plant conservation projects

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In the frame of Seedforce project (LIFE20 NAT/IT/001468), which aims to improve the conservation status of 29 endangered native plant species in Italy, Malta, France and Slovenia. Assessing genetic diversity is an essential aspect while defining a specific conservation strategy, as it provides insights into the variability of donor populations. Minimizing the selection of closely related individuals reduces the risk of inbreeding, while maintaining genetic variability ensures the adaptability and long-term survival of the translocated populations.

For none of the project species a reference genome was available. Thus, the genotyping method of choice was ddRAD-seq, a method enabling genotyping thousands of loci in species for which no extensive sequence information is available.

With the aim of optimizing sequencing resources, most of the species were sequenced after pooling DNA from eight individuals, and information on expected heterozygosity and beta diversity were obtained. For six of the project species, samples were sequenced individually, which allowed to perform expected heterozygosity and beta diversity analyses, information on observed heterozygosity, kinship, inbreeding and population structure. Approximately 500 samples were sequenced individually, and 800 were sequenced as 100 pools of eight individuals each. One pipeline for individual approach and another for pooled individuals were created.

Our results showed that overall, for the species that were sequenced following the individual approach, heterozygosity was low, ranging from 10% to 4% with Inbreeding values higher than zero in all the species. In many situations inbreeding coefficient was around 0.2, with the notable exception of one taxon (*Linaria flava* subsp. *sardoa*), in which inbreeding coefficient was higher than 0.9. The individual approach also showed high inter-population differentiation within the same species. The pattern of low genetic diversity and high isolation is compatible with the expectation for small, fragmented populations.

The pooled approach revealed low to moderate expected heterozygosity, ranging from 10% to 35%, with high population differentiation, again supporting the expectation for small, fragmented populations. Information on genetic diversity of the populations of each species will be used to inform the translocation policies foreseen in the project.

TERSICORE Project: a multidimensional approach to urban greening, environmental monitoring, and community engagement for Urban Regeneration.

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Green spaces in urban areas play a fundamental role in shaping the cityscape, providing critical benefits in mitigating the effects of extreme weather events, such as heatwaves, flooding, and droughts, as well as urban pollution. Beyond their environmental impact, these spaces also contribute to the psychological well-being of individuals by fostering place attachment, reducing stress, and promoting a sense of community.

In this context, the TERSICORE (Technologies for Evaluation and Research for Sustainability Innovation: a Community-Oriented Regenerative Evolutions) project fits within the broader framework of urban regeneration, employing a multifaceted and multidimensional approach to improve urban environments through sustainable, inclusive, and innovative practices. The project encompasses an architectural dimension, consisting of the design and construction of modular, flexible, and interactive installations tailored to transform micro-urban spaces into collective gathering areas, as well as a psycho-social dimension, integrated through practices of self-construction and active community engagement, aiming to transform marginal spaces into areas of social cohesion and interaction.

The project is also featured by an environmental dimension focusing on the health of the urban ecosystem and the crucial role of green spaces. This dimension involves targeted greening interventions, guided by the evaluation of environmental quality in accordance with relevant UN-Habitat guidelines collected through the Urban Indicator Database (<https://data.unhabitat.org/>). The selected indicators, chosen based on the characteristics of the intervention area, align with the sustainability criteria and Sustainable Development Goals (SDGs) identified by the 2030 Agenda. This analysis included assessing the quantity and quality of urban green areas and their contribution to improving urban livability through enhanced environmental comfort. Greening interventions will be implemented by the installation of biosensors, able to monitor air quality in urban spaces by the moss-bag technique, a well-established method based on mosses' capacity to adsorb and accumulate atmospheric pollutants such as heavy metals, particulate matter (PM10 and PM2.5), and volatile organic compounds (VOCs). This provides an effective, cheap and versatile system for monitoring urban air quality.

Environmental and social analyses will be conducted before, during, and after the interventions, offering a valuable opportunity to evaluate the effectiveness of urban regeneration and greening strategies implemented by the project. This multidisciplinary and integrated approach has been tested as a case study within the Sa Duchessa complex at the University of Cagliari, enabling the development of a reproducible and standardized protocol while also identifying key limitations and critical aspects essential for effective planning and implementation at a larger scale. Furthermore, the project includes direct application beyond the university context, with an upcoming implementation scheduled in the urban area of Cagliari.

In conclusion, TERSICORE aims to establish and strengthen micro-systems of green infrastructure with potential effects on various ecosystem services and urban biodiversity. The project seeks to develop low-cost, rapid, and adaptable solutions that can provide direct insights to urban planning policies, promoting participatory approaches to urban transformation.

Modelling seed germination data to meet conservation needs in Mediterranean habitats

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Seed germination knowledge under changing environments is critical to support effective seed-based solutions for sustainable development. While germination data archived in open-access repositories can help synthesise broad-scale germination patterns, their adequacy for fine-scale modelling remains largely unexplored.

Here, we assessed the current availability and quality of germination data for a species pool to meet conservation needs in Mediterranean habitats. We hypothesised that: (1) germination data are limited for species that exhibit vegetative, reproductive and seed traits that hamper seed collection, processing, and germination; and data quantity and quality is not a constraint for (2) species with wide ecological breadth, (3) conservation species, or whose seeds are commercially available for restoration or have reported uses. We tested these hypotheses using phylogenetic logistic regressions and further assessed the adequacy of the available data by modelling the species' seed germination thermal responses using quadratic regressions.

Overall, germination data were available for 31% of the species, yet only 9% had adequate data for thermal modelling. Data were more abundant and adequate for thermal modelling in species with wide ecological breadth, setting indehiscent fruits, physiological dormant seeds, commercially available for restoration, and reported plant uses. Surprisingly, despite limited data availability, perennial herbs and species for conservation interest had adequate data for germination thermal modelling. Our study highlights key gaps in open-access repositories, offering a framework for future data collection and analysis to enhance conservation and restoration efforts. Furthermore, our results provide novel insights into the thermal tolerances and germination risks faced by Mediterranean species. Species exhibiting complex dormancy mechanisms and cold, narrow thermal niches are particularly vulnerable to climate change, as shifts in temperature and seasonality may desynchronise germination cues and compromise recruitment. These findings highlight the urgent need to prioritise data collection and germination research for underrepresented taxa and to promote the use of species with lower thermal risk profiles in restoration planning, ensuring effective conservation strategies under future climate scenarios.

The BRIGANTINE project: a new sustainable approach for monitor the health status of seaweeds and seagrasses in the North Adriatic Sea

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Coastal areas include some of the most threatened natural ecosystems worldwide. Indeed, habitat degradation, environmental pollution and climate change are altering coastal systems, causing an important loss of ecological services. Amongst coastal ecosystems, algal forests and seagrass meadows (AFSMs) provide food and shelter for a wide variety of organisms, recycle nutrients and protect shoreline from erosion (1). Given the great importance of these communities, their continued monitoring based on multiple indicators is essential to assess their health status and their responses to anthropogenic impacts. In this context, hyperspectral imaging is playing an increasingly important role. This approach has been efficiently applied for remote and long-term monitoring of ecosystems, including coastal environments (2), which introduce additional challenges compared to terrestrial habitats due to the attenuation of the visible and infrared radiation.

The Interreg BRIGANTINE project aims at monitoring the health status of AFSMs distributed in the North Adriatic Sea developing an Automated Surface Vehicle (ASV) (Fig. 1) equipped with multispectral cameras and chemico-physical sensors. The captured images will be used for the detection of the spectral properties of the macrophytes, in order to apply spectral indices and assess their health status (Fig. 2). Although non-destructive monitoring stands as one of the main advantages of remote sensing, validating the meaning of the spectral signature with field physiological and morphological traits considered good indicators of the health status of these macrophytes is important to understand the monitoring potential of such tools. Specifically, pigments (i.e., chlorophyll *a*, *b* and carotenoids), phenolic compounds and carbohydrates and several morphometric variables, will be taken into account due to their significative relation with abiotic stresses. Furthermore, chemico-physical parameters (i.e., metal ions and macronutrients) measurement in the water column will provide a real-time information about the possible stresses faced by AFSMs. The development this novel monitoring approach of AFSMs will provide crucial information on the health status of these communities, valuable to plan measures for the preservation and management of these fragile ecosystems.



Fig. 1. Automated Surface Vehicle equipped with multispectral cameras and chemico-physical sensors.

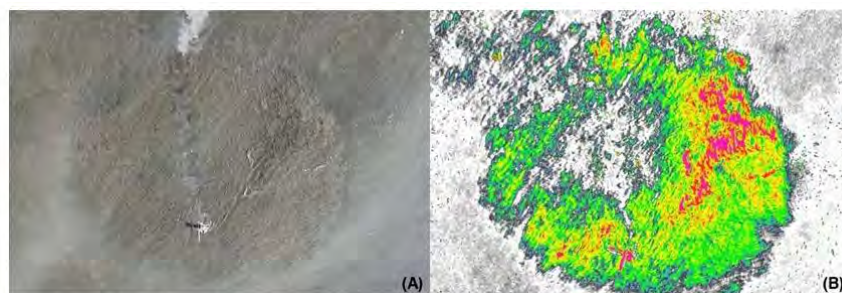


Fig. 2. Drone images of a seagrasses patch in the Marano Lagoon (Italy). (A) True colors image and (B) multispectral image.

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The mycobiome of the critically endangered *Abies nebrodensis*: environmental conditions are the drivers of fungal diversity

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The Sicilian fir (*Abies nebrodensis*), a critically endangered endemic species of northern Sicily, survives as a relict population of approximately 30 adult trees within the Madonie Regional Park (Italy). Its persistence in a fragmented habitat characterized by harsh and variable ecological conditions makes it particularly vulnerable to environmental stressors and biotic threats.

The preservation of this species has been faced by the EU project “LIFE4FIR” (LIFE18/NAT/IT/000164, 2019–2023), aiming at the conservation of *A. nebrodensis* by both in situ and ex situ conservation actions.

The study presented here focused on the characterization of the phyllosphere mycobiome of *A. nebrodensis*, to explore how environmental context, plant health, and organ type influence fungal diversity and community structure. High-throughput sequencing (Illumina MiSeq, ITS2 region) was used to analyze 80 pooled samples collected from symptomatic and asymptomatic needles and twigs across three distinct ecological clusters.

The results revealed a complex and diverse fungal assemblage, with 4567 Amplicon Sequence Variants (ASVs) identified, of which nearly 48% were annotated to species level. Alpha and beta diversity analyses indicated that the total fungal community (epiphytic + endophytic) exhibited significantly higher richness and compositional variability than the strictly endophytic fraction. Notably, ecological variability had a stronger influence on fungal community structure than either plant health status or organ type. Trees located in the most exposed and nutrient-poor sites harbored fungal communities that were compositionally distinct and less diverse.

Taxonomic analysis highlighted the prevalence of several fungal families, such as Valsaceae and Phaeosphaeriaceae, in both symptomatic and healthy tissues. Functional profiling suggested the coexistence of saprotrophs, pathogens, and mutualists, with no strong evidence of specific taxa driving disease symptoms. A core microbiota was identified for both the total and endophytic communities, with *Phaeosphaeria* emerging as a dominant and consistently present genus.

These findings suggest that fungal communities associated with *A. nebrodensis* are shaped predominantly by environmental conditions rather than host health alone. Understanding these microbiome dynamics provides important insights for the conservation and management of this endangered conifer under changing climatic scenarios.



Fig. 1. *Abies nebrodensis* in its ecological area

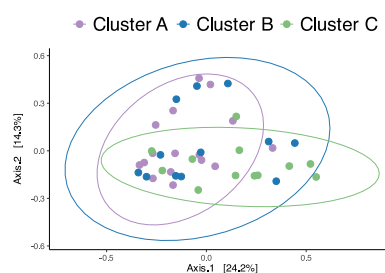


Fig. 2. Bray-Curtis non phylogenetic beta-diversity of the phyllosphere total fungal communities of the three ecological clusters

Hotspots of Alpine Vascular Plants in the Andes and Their Conservation Status

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The Andes, the longest mountain range in the world, harbour an exceptionally high number of endemic plant and animal species, which are increasingly threatened by ongoing climate change, land-use and habitat fragmentation (1, 2). Recent analyses, based on protected area data from the World Database on Protected Areas (WDPA), indicate that only 15.8% of the Andes are protected, which is significantly below the 30% conservation target set by the Kunming-Montreal Global Biodiversity Framework. This study aims to delineate alpine plant biodiversity hotspots across the Andes and evaluate their current conservation status (3).

Occurrence data for all alpine vascular plant species of the Andes were obtained from GBIF, resulting in 107,410 georeferenced records within the Andean region, as defined by the GMBA Mountain Inventory v2.0 (Fig. 1a). The study area was divided into 50 × 50 km cells, and four indices (species richness, endemic richness, range-restricted endemic richness, and range rarity richness) were calculated. Cells were ranked and grouped into richness percentiles. A grid cell qualified as a hotspot if it fell within the uppermost 5%, 5–10%, and 10–20% richest cells for at least one index (Fig. 1b). Protected areas were integrated using the WDPA data (IUCN categories I–IV). A hotspot was classified as a conservation gap if fewer than 10% of its area was covered by protected areas.

From a total of 1,459 alpine species, 1,370 were endemic to the Andes (94%), and 585 species were range-restricted, i.e., occurring in less than 1% of the study area (43% of the endemics).

A total of 70 cells were identified as top 5% hotspots, 101 cells as top 5–10% hotspots, and 149 cells as top 10–20% hotspots. These hotspots were mostly found in the Central Andean Puna, northern Andean Páramos, and Bolivian and Peruvian Yungas ecoregions. However, only 27%, 23%, and 20% of the top 5%, 5–10%, and 10–20% hotspots, respectively, are currently protected, while the remaining areas represent conservation gaps (Fig. 1c). These patterns highlight significant conservation gaps in the Andes, emphasizing the urgent need to enhance protection efforts in these biodiversity-rich regions.

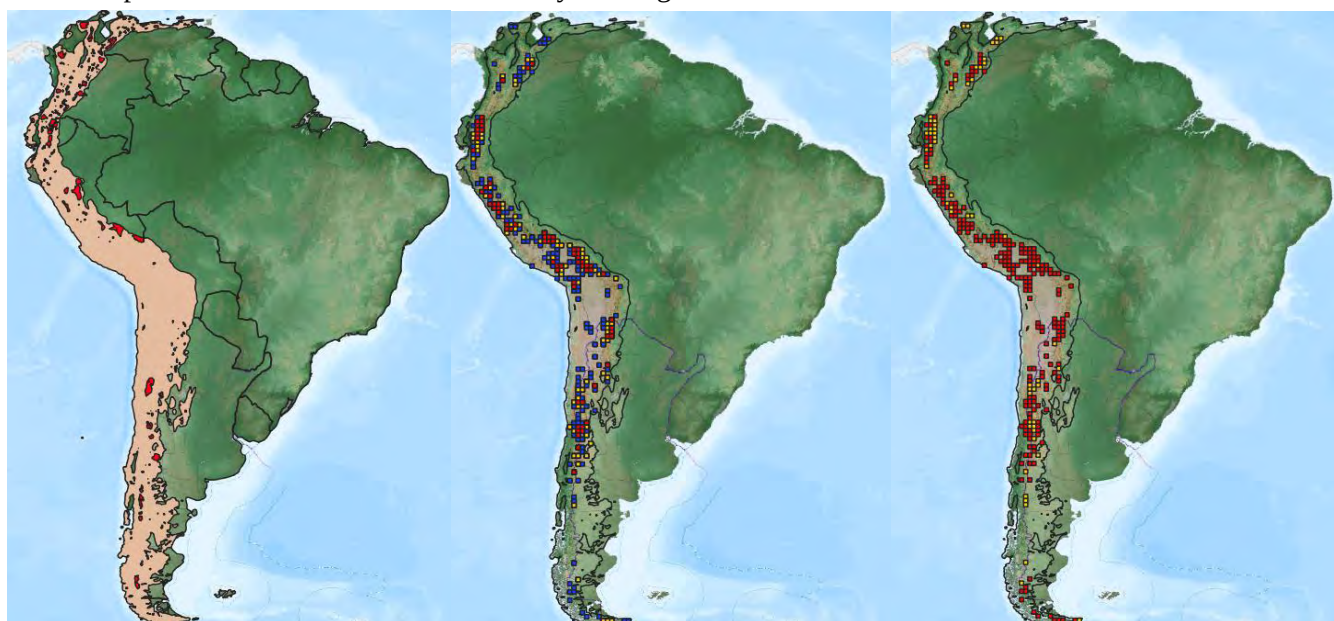


Fig. 1a. Study area. Andean region defined by GMBA Mountain Inventory v2.0 (brown) with IUCN areas I–IV (red).

Fig. 1b. Hotspots of alpine plant diversity in the Andes: top 5% (red), 5–10% (yellow), and 10–20% (blue) hotspots.

Fig. 1c. Top 20% richest biodiversity cells. Less than 10% protected (red), more than 10% protected (yellow).

1) Myers, N. et al. (2000) 'Biodiversity hotspots for conservation priorities', *Nature*, 403(6772), pp. 853–858. Available at: <https://doi.org/10.1038/35002501>

2) Cuesta, F. et al. (2017) 'Latitudinal and altitudinal patterns of plant community diversity on mountain summits across the tropical Andes', *Ecography*, 40(12), pp. 1381–1394. Available at: <https://doi.org/10.1111/ecog.02567>

3) Theobald, D.M. et al. (2024) 'Evaluating ecosystem protection and fragmentation of the world's major mountain regions', *Conservation Biology*, 38(3), p. e14240. Available at: <https://doi.org/10.1111/cobi.14240>

Biomonitoring of potentially toxic elements in the Venice Lagoon (Italy): a 100-year comparison using museum specimens

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The Venice Lagoon is a unique yet vulnerable ecosystem, hosting remarkable biodiversity while undergoing significant anthropogenic pressures and morphological alterations, especially throughout the 20th century. During the 1930s, Aristocle Vatova established a comprehensive baseline through systematic surveys of macroalgal diversity, collecting specimens from around 70 sites. These samples - taxonomically validated by Victor Schiffner and preserved at the Natural History Museum of Venice (1)- provide a valuable historical record of algal biodiversity (1930-1932) and offer unique opportunities for utilizing Natural History Collections (NHCs) in for interdisciplinary environmental research, including long-term biogeochemical studies.

To assess the long-term changes in elemental composition, we conducted a comparative analysis between these historical specimens and contemporary *Ulva* spp. samples collected in 2025. From the Vatova survey, we selected 22 sites where at least 10 specimens of *Ulva* were recorded. In May 2025, we resurveyed these sites (or nearby alternatives where access was restricted). For each sampling site, ≥ 10 thalli were collected, in order to ensure sufficient replicates for analysis. Collected samples were oven-dried, homogenized, and processed (≥ 1 g dry weight per replicate). Due to the non-destructive nature of then investigation required for museum specimens, elemental analysis was performed using portable X-ray fluorescence (XRF) spectroscopy.

Our study provides a quantitative century-long record of potentially toxic element accumulation in *Ulva* spp., the lagoon's dominant macroalgal taxa, revealing significant temporal patterns in anthropogenic impact. Furthermore, we demonstrate the crucial value of NHCs as archives for environmental change studies and present a transferable methodological framework for historical-ecological investigations in coastal ecosystems.

1) L. Seggi, R. Trabucco, S. Martellos (2024) *Plants*, 13, 2901. <https://doi.org/10.3390/plants13202901>

Decline of High Nature Value Grassland Habitats Within Natura 2000 Sites in Slovenia

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Grasslands in Slovenia are an important element of the agricultural landscape and one of the most species-rich habitats in Europe. Despite their ecological and agricultural value, a decline in species-rich grasslands has been observed over the last two decades, even within Natura 2000 protected areas. Our study explores the loss of High Nature Value (HNV) grasslands since Slovenia's admission to the EU and inclusion in the Natura 2000 network in 2004.

To quantify the loss of grassland, we selected 33 Natura 2000 sites for which habitat mapping data were available. Using GIS tools, we compared older habitat type layers (from 2000–2018) with land-use data from the 2023 national cadastre of agricultural land. Habitat types were cohered by translating PHYSIS codes (classification of Palearctic habitats) into FFH (Natura 2000) classifications. We focussed on ecologically important semi-natural grassland types and used spatial intersection analysis in QGIS to determine where formerly mapped grasslands had been converted to non-grassland land use.

The analysis revealed an overall 25 % decline in grassland area across all studied sites, with local losses varying widely - from as low as 6 % to over 50 %. Some of the most affected areas were Ljubljansko barje (–27 %), Goričko (–39 %) and Ribniška dolina (–49 %). The most heavily impacted habitat type was 6510 - lowland hay meadows (*Arrhenatherum elatius*, *Alopecurus pratensis*), which were often converted to arable land or abandoned and overgrown. In a few cases (e.g. Planinsko polje), where more habitat mappings are available, we also observed a dramatic degradation in habitat quality, such as a 55% decline in habitat type 6410 (*Molinion caeruleae*) within 13 years.

Our results are preliminary and have some limitations due to inconsistency in mapping years. However, they highlight the systematic decline of grasslands within protected areas and emphasise the need for better coordinated nature conservation policies, improved monitoring and investment in conservation and restoration measures.

Monitoring of wild orchids in urban and natural ecosystems and their potential pollinators

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Orchids (Orchidaceae family) are plants that can grow in many environments and are distributed in almost all terrestrial habitats, showing the highest species diversity in tropical regions. They are amazing plants with definite ecological requirements: in fact, they establish a specific association with mycorrhizal fungi during different phases of their life cycle, which allows plants to acquire nutritive compounds, in a fascinating and unique symbiotic relationship. Interactions orchid-fungus/i mainly influence the establishment, size and distribution of orchid populations, addressing spatio-temporal variations, dynamics of plant communities and biodiversity (1). Furthermore, orchids have evolved outstanding relationships with pollinators through different attraction strategies; consequently, they are considered plants with a high grade of specialisation for their pollinators. In fact, about 60% of orchids appear to be pollinated by only a single pollinator species or a very restricted number (2).

Monitoring wild flora can sometimes reserve pleasant discoveries. Despite anthropic disturbance, pollution and invasion of alien species, in recent years some orchid species have been observed in urban areas, and their number appears to be increasing, as shown by frequent reports coming from researchers, specialists of local flora and citizens. We started some years ago to collect data on orchid occurrence in the cities of Modena and Reggio Emilia with the aim of studying, monitoring and promoting conservation actions. *Himantoglossum adriaticum* was first observed in 2019 in a public park in Reggio Emilia (Parco S. Lazzaro) on a small lawn regularly mowed. Then, the number of individuals in different places, but within the same area, is gradually increased, reaching 12 flowering plants in 2025 and, in addition, a lot of other young plants showing only rosettes. Its occurrence is notable as this species has been included in Annex II Directive 92/43/EEC (Habitat Directive); it has recently been assessed for The IUCN Red List of Threatened Species (2010) and is actually listed as Least Concern. It is considered a wide-spread food-deceptive orchid and has recently been classified as functionally generalised for pollinators (3).

The evaluation of the rate between the number of flowers and the number of capsules produced for each plant has been used as an indicator of reproductive efficiency and indirectly of the pollinator's occurrence. In the area of study, *Himantoglossum adriaticum* plants showed a good production of capsules, 41.67% (2023), 68,13% (2024), considering the average percentage between all plants. An experiment was set up to verify whether insects could contribute to the pollination of *H. adriaticum*. The experiment compared the visitors observed in an urban ecosystem (the above-described public park in Reggio Emilia) and a natural reserve (Salse di Nirano in the province of Modena). Before anthesis, nine plants were selected at each site: six of these were hooded with fine nylon mesh to prevent insects from entering the flowers, while three were left uncovered. Of the six hooded orchids, two were kept closed until the end of flowering (to act as a control for self-pollination), and four were uncovered for each 30-minute sampling session, over three consecutive weeks. During this time, they were filmed with micro-cameras. Meanwhile, one operator took pictures of insects entering the flowers, while another was ready to catch them (placing them in sterile vials) as soon as they left the plants. The videos, photos and insect samples were then carefully examined to identify the species and check for the occurrence of orchid pollinia. Preliminary results showed that a greater number of insect visitors and a higher level of diversity were present in the natural reserve. Several species belonging to the Anthophila (Hymenoptera), particularly the Halictinae and Andreninae subfamilies, as well as some very small Diptera, were captured at both sites. The precise identification of the insect specimens and detection of orchid pollen are ongoing.

1) T. Li, S. Wu, W Yang, M.A. Selosse, J. Gao (2021) Front. Plant Sci., 12, 647114

2) R.L. Tremblay (1992) Can. J. Bot., 70, 642-650

3) E. Fantinato, S. Del Vecchio, M. Baltieri, B. Fabris, G. Buffa (2017) Ecol. Res., 32, 951-959

Advanced Monitoring of Phytoremediation Effectiveness in Multi-Contaminated Sites through Metaproteomic approach

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Phytoremediation is emerging as a sustainable and eco-compatible approach for the reclamation of contaminated soils, particularly when integrated with biotechnological strategies involving microbial consortia inoculation, establishment of plant species and soil amendments. However, the validation of these approaches under real conditions requires long-term field studies capable of capturing environmental complexity and microbial functional responses. In this study, we investigated the efficacy of a five-year phytoremediation trial conducted on a multi-contaminated site, integrating high-resolution metaproteomics to assess microbial dynamics and functional shifts. A key aspect of this work was the use of an optimized protein extraction protocol specifically tailored for contaminated soils, which enabled high-yield recovery and robust proteome coverage. Significant reductions in contaminant concentrations were observed in the treated plots (A1, A3, A7), with decreases exceeding 60% for heavy metals, PAHs, and PCBs, whereas control soils (Bulk) showed negligible variation. Metaproteomic analysis revealed clear differences between treated and untreated soils, both in terms of taxonomic composition and protein expression profiles. In the treated areas, there was a pronounced increase in proteins associated with central carbon metabolism—including glycolytic enzymes such as glyceraldehyde-3-phosphate dehydrogenase (GAPDH), enolase, and fructose-bisphosphate aldolase—as well as key tricarboxylic acid (TCA) cycle enzymes like malate dehydrogenase and isocitrate dehydrogenase. This was accompanied by the upregulation of ATP synthase subunits and enzymes involved in anabolic pathways, indicating an active and functionally restored microbiome. Moreover, a marked abundance of molecular chaperones (e.g., GroEL, HSP70, HSP90) in treated soils suggested cellular adaptation to residual stress and ongoing microbial activity. In contrast, control soils exhibited proteomic signatures dominated by stress response proteins and oxidative defense enzymes such as superoxide dismutase and catalase, indicative of persistent environmental pressure. Taxonomically, the treated areas showed an enrichment of microbial taxa known for their pollutant-degrading and metal-tolerant capacities, including *Sorangium cellulosum*, *Pseudomonas knackmussii* and *Microbacterium hydrocarbonoxydans* for PAHs and PCBs, and *Thiohalomonas denitrificans*, *Tumebacillus flagellatus* and *Kineococcus radiotolerans* for heavy metals. These findings were consistent with the improved chemical profiles and reflected a restructured and functionally resilient soil microbiome. Overall, the integration of metaproteomics in long-term field experimentation provided direct evidence of microbial functional restoration and effective contaminant degradation. The observed enzymatic and taxonomic shifts not only confirmed the success of the phytoremediation approach but also highlighted the utility of proteomic biomarkers in evaluating soil recovery. This study underscores the importance of combining molecular-level insights with environmental monitoring to inform the development of reliable protocols for large-scale soil bioremediation.

POSTERS

1.3 = The effects of land-use change on native and alien species in the Conca d'Oro of Palermo (Sicily)

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The Conca d'Oro of Palermo, a historically and agriculturally vital plain in northwestern Sicily, has experienced substantial landscape changes due to intensified farming and urban expansion. The plain spans approximately 100 km², with a gentle slope descending from 100 meters above sea level at the mountain foothills to the coast.

The Conca d'Oro was once abundant with freshwater springs that emerged at the base of the mountains, feeding streams and creating extensive wetlands. Over time, these spring-fed marshes were drained and repurposed, while the lower river sections were straightened and their beds cemented. Additionally, the brackish Mondello wetlands were reclaimed in the early 20th century.

The site of modern-day Palermo has been inhabited since prehistoric times and has witnessed at least twelve successive ruling civilizations. Each of these cultures left its mark on the city's evolution and its surrounding environment—not merely through architectural contributions, but more significantly through the systematic exploitation of local resources. Before human settlement, the area was covered by dense, continuous vegetation. During Roman rule (254 BC-440 AD), extensive holm oak forests were cleared for cereal production. The Arab introduction of irrigation in the 9th century subsequently diminished both cereal fields and uncultivated lands. Major 16th century land reclamation projects eliminated wetland ecosystems, while the early 1800s saw the rise of intensive citrus cultivation (particularly lemons and mandarins). Most recently, post-World War II urban development has consumed significant agricultural land.

The landscape has undergone a continuous evolution since 1750, marked by distinct phases of transformation: first from semi-natural to agricultural land, then subsequently to urbanized areas. Palermo's urban core remained contained within its historic walls until the early 20th century, experiencing particularly rapid expansion in the post-World War II period.

A total of 40 plant taxa once documented in the Conca d'Oro have now vanished from the area. Most of these were associated with wetland habitats or coastal dunes, while a smaller number belonged to cultivated and uncultivated drylands. The most severe habitat losses occurred in the Mondello marshes, the Oreto River estuary wetlands, and the San Ciro and Baida marsh systems.

Several species, including *Descurainia sophia* (L.) Webb ex Prantl, *Elatine macropoda* Guss., and *Stuckenia pectinata* (L.) Börner, once common along canals and watered fields, disappeared due to shifts in farming practices that eliminated the irrigation ditches they depended on. Another group of locally extinct species includes those of coastal sandy habitats such as *Cressa cretica* L., *Echium arenarium* Guss., and *Sporobolus pungens* (Schreb.) Kunth.

The introduction and establishment of non-native species has followed distinct historical pathways. Archaeophytes, accompanying agricultural development, have gradually naturalized into the vegetation over centuries. In contrast, many neophytes, initially introduced as ornamental plants, have demonstrated invasive tendencies, aggressively colonizing areas where native vegetation had been degraded or removed. Similarly some native species such as *Ceratonia siliqua* L., *Myrtus communis* L., and *Viburnum tinus* L. have been extensively cultivated and now frequently occur in semi-natural habitats.

This study highlights both the value and limitations of archival materials for reconstructing urban landscape evolution and floristic changes, while advocating for the protection of remnant ecological refuges - particularly rugged features like coastal bluffs and riparian gorges. These biodiverse microhabitats not only preserve threatened flora and fauna but also provide urban residents with vital connections to natural systems.

Project funded under the National Recovery and Resilience Plan (NRRP), Mission 4, Component 2, Investment 1.4-Call for tender No. 3138 of 16 December 2021, rectified by Decree n. 3175 of 18 December 2021 of Italian Ministry of University and Research funded by the European Union - NextGenerationEU Project Code CN_00000033, Concession Decree No. 1034 of 17 June 2022 adopted by the Italian Ministry of University and Research, CUP B73C22000790001, Project Title "National Biodiversity Future Center-NBFC.

1.3 = A Contribution to the Lichen Biota of Armenia

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The lichen biota of Armenia currently comprises 619 (1,2,3) taxa. The aim of this study was to update the current lichen checklist of Armenia with new records. Lichens and lichenicolous fungi were collected at eleven localities, on different substrates and altitudes, from lowland areas to the alpine/subalpine belts. A total of 144 taxa, including 131 lichens, nine lichenicolous lichens and four lichenicolous fungi were identified. Of these, 46 species are new to Armenia, which highlights the need to further investigate lichen diversity in the country to establish a more complete checklist. Even common or very characteristic species turned out to be new for the country, e.g., *Verrucula biatorinaria* (Fig. 4) and *V. pusillaria* (Fig. 2), two lichenicolous lichens growing on *Calogaya biatorina* and *C. pusilla*, respectively. This result is quite surprising, as both *Calogaya* species are widespread in Armenia and almost all the samples collected by us were parasitized. Less common species, e.g., *Arthopyrenia parolinii* (Fig. 1) and *Protoparmeliopsis usbekica* (Fig. 3), were previously known only from a few historical records in Veneto (Italy) and from the most continental regions of Asia, respectively. These may be often overlooked because of their small size (*A. parolinii*) and different major distribution (*P. usbekica*). Furthermore, to explore the variation and the diversity of photobionts in alpine lichens, we selected lichen species that were collected at least at two different altitudes along an altitudinal transect on Mt. Aragats. From each species and the selected thalli, the photobionts were isolated in culture and identified by molecular and morphological characters.



Fig. 1. *Arthopyrenia parolinii*



Fig. 2. *Verrucula pusillaria*

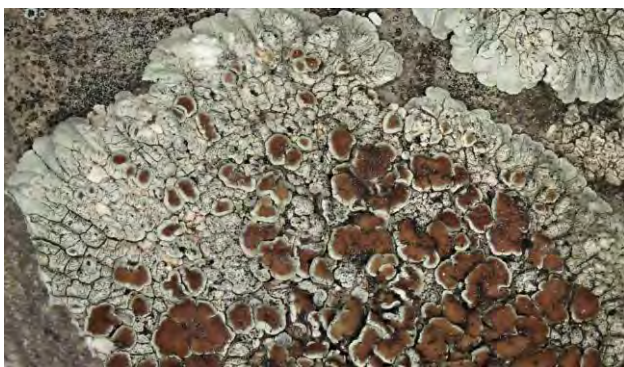


Fig. 3. *Protoparmeliopsis usbekica*

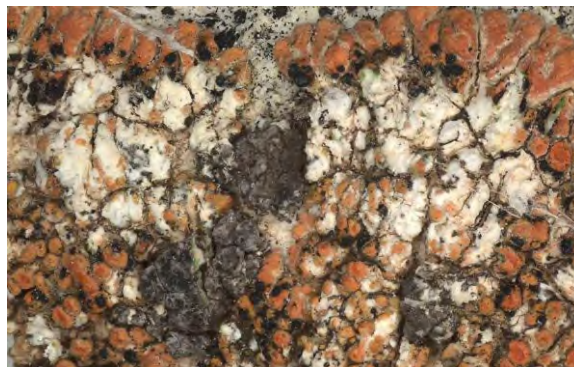


Fig. 4. *Verrucula biatorinaria*

The scale bar is equivalent to 1mm.

1) A. Gasparyan, H. Sipman (2016) Phytotaxa, 281, 1-68

2) A. Gasparyan, H. Sipman (2020) Herzogia, 33, 554-55

3) A. Gasparyan, H. Sipman, R. Lücking (2017) The Lichenologist, 49(4), 301-319

1.4 = What do fossil Charophytes whisper to us? Palaeoecological and palaeoenvironmental reports from Pleistocene continental deposits of Umbria (Central Italy)

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Fossil charophyte assemblages from the Early Pleistocene deposits of the Tiberino Basin (Central Italy) offer a valuable record for reconstructing palaeoenvironmental dynamics during a climatically pivotal epoch in the Mediterranean region. This study integrates micropalaeontological and sedimentological analyses from three key sites—Arquata Quarry (Bevagna), Ellera di Corciano, and Acquasparta Well (Fig. 1)—to interpret ecological signals embedded within charophyte remains. The dominant taxa identified include *Nitellopsis obtusa* (Desv.) J.Groves, *Lychnothamnus barbatus* (L.Meyen) Leonh., *Chara hispida* L., *Chara vulgaris* L., and *Lamprothamnium papulosum* (Wallr.) J.Groves. These assemblages represent a spectrum of freshwater environments ranging from deep, oligotrophic lakes to ephemeral, carbonate-rich wetlands. At Arquata, the co-dominance of *N. obtusa* and *L. barbatus* suggests prolonged lacustrine stability during humid intervals. In contrast, the prevalence of *L. papulosum* in Ellera and Acquasparta indicates fluctuating salinity and transient hydroperiods—hallmarks of heightened aridity and climatic seasonality during the Early–Middle Pleistocene transition (~1.4 Ma). Particular emphasis is placed on the palaeolimnological significance of *L. papulosum*, a species well-adapted to variable hydrological conditions and a reliable ecological proxy for hydroclimatic instability. Furthermore, travertine deposits at Acquasparta record episodes of groundwater resurgence under semi-arid climatic conditions. Comparative analysis with contemporaneous Mediterranean sites—such as Laguna de Gallocanta (Spain) and Lake Afourgagh (Morocco)—corroborates these palaeoecological interpretations, affirming their reflection of broader regional climatic patterns. The findings highlight the sensitivity of charophyte communities to environmental fluctuations and reinforce their utility in Quaternary palaeohydrological reconstructions. This research contributes to a deeper understanding of continental basin evolution in the Mediterranean and underscores the palaeoenvironmental value of charophyte gyrogonites.

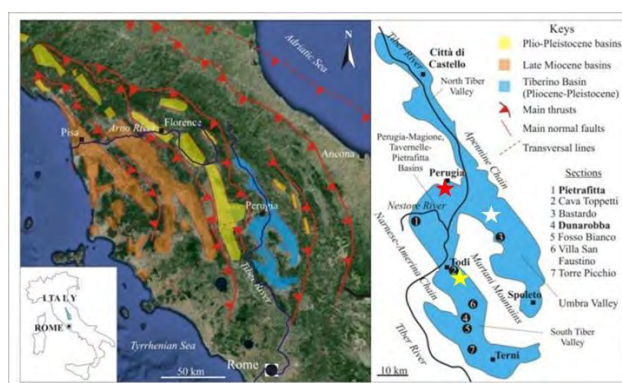


Fig. 1. Map showing the distribution of the main sedimentary basins in Central Italy and a stratigraphic profile of the Tiberino Basin, indicating the locations of the studied successions: Arquata Quarry (white star), Ellera di Corciano (red star), and Acquasparta Well (yellow star), modified from Bizzarri et al. (2018)

1) R. Bizzarri, P. Corrado, D. Magri, E. Martinetto, D. Esu, V. Caprai, R. Colacicchi, G. Napoleone, A. Albianelli, A. Baldanza (2018) *Quaternary Research*, 90 (1), 201-221

2) A. Rey-Boissezon, D. Auderset Joye (2015) *Aquatic Botany*, 120(A), 84-91

3) M. Cherin, R. Bizzarri, N. Buratti, T. Caponi, F. Grossi, T. Kotsakis, L. Pandolfi, F. Pazzaglia, M.R. Barchi (2012). *Rend. Online Soc. Geol. It.*, 21, 1075-1077

1.5 = Clonal growth diversity in semi-dry calcareous grasslands (Slovenia)

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Given the widespread importance of clonality in plants, investigating the mechanisms by which clonality-related traits influence community organisation is crucial for a functional understanding of vegetation. In this study, we examined the patterns of clonality and other life-history traits in a series of species-rich semi-dry grasslands (alliance *Bromion erecti*, order *Brometalia erecti*, class *Festuco-Brometeta*) in the Central European region of Slovenia, all assigned to the same EUNIS (Middle European *Bromus erectus* semi-dry grasslands, code R1A32) and Natura 2000 habitat type (code 6210*, Semi-natural dry grasslands and scrubland facies on calcareous substrates (*Festuco-Brometalia*) (* important orchid sites). The objectives of our research were (i) to assess the diversity of clonal growth organs (CGOs) and other clonal traits in semi-dry grasslands and (ii) to determine the differences in the distribution of CGOs and other clonal traits between three different plant communities (associations) in the area. We analysed data from 224 vegetation plots (phytosociological relevés) containing 247 species, examining nine plant traits of grassland species. We found that over half of the plant species were clonal. The most common clonal growth organs (CGOs) were the perennial splitting main root (produced by non-clonal plants), epigeogenous rhizomes and hypogeogenous rhizomes. The community weighted mean (CWM) of all plant traits was calculated for each plot to determine differences between the three grassland types. Soil moisture and productivity appear to be the key factors determining differences in clonal growth between associations. Clonal tussock plants with rhizomes and species with a richer bud bank and a higher number of clonal offspring were abundant in grasslands with deeper soils and relatively mesic conditions (the *Onobrychido-Brometum* association). In contrast, grasslands with stony soils over fissured limestone or dolomite, and with drier conditions, were characterised by rosette plants with hypogeogenous rhizomes (*Bromo-Danthonietum calycinae*) and/or non-clonal plants with splitting main root (*Scabioso hladnikianae-Caricetum humilis*). The diversity and characteristics of clonality-related traits indicate that clonal growth is an essential feature of these species-rich grasslands. Our results suggest that management plans for semi-dry grassland habitats in the study area should be designed at a finer scale than at the level of EUNIS and Natura 2000 habitat types, for example at the level of associations, in order to support the characteristic species and functional composition of all grassland types.

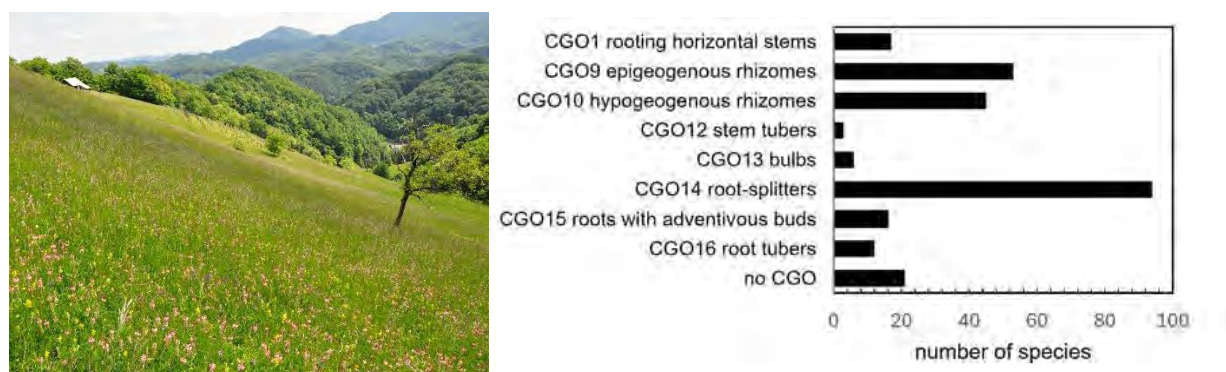


Fig. 1a. Orchids-rich *Bromion erecti* meadow (Annex I habitat type, code 6210*) in hilly region of NE Slovenia (photo S. Škornik); 1b. distribution for types of clonal growth organs (CGOs) for 247 plant species in dry grasslands (Natura 2000 habitat type 6210*) in Central European part of Slovenia

1.8 = Intraspecific leaf trait variation, genetic structure and adaptive strategies in forest endemics: a case study from the genus *Aegonychon* (Boraginaceae)

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The Italian flora includes approximately 1700 endemic plant species, around 130 of which are found in woodlands and forests of the South and the Islands (1). Most of these endemics are herbaceous understory species, which account for up to 80% of the biodiversity in temperate forests and play key ecosystem roles, such as the cycling of carbon and other elements. Several historical, ecological, or biological factors may contribute to the emergence and development of endemic taxa, including a relatively recent origin, a narrow ecological niche, limited dispersal ability, low adaptive phenotypic plasticity, or genetic variability. As part of a project on Italian forest endemics, we aimed to assess the role of these factors using a model system consisting of a species pair with one narrow-range endemic and one closely related species with a broad continental distribution. These species are *Aegonychon calabrum* (Ten.) Holub and *A. purpureocaeruleum* (L.) Holub, belonging to the tribe Lithospermeae in family Boraginaceae (2). The former occurs with scattered populations in mountain forests of southern Italy, whereas the latter is widespread in the understory of thermophilic forests across many European countries and in all Italian regions, except Sardinia. We analysed representative population samples of both species to evaluate potential differences in the range and patterns of intraspecific variation in key leaf traits associated with resource-use attitudes (LA, SLA, LDMC), as well as in Grime's ecological strategies and genetic structure. We also compared genetic parameters and phenotypic traits to evaluate the roles of genetic drift and natural selection at the intraspecific level. LA and LDMC showed significant differences, with higher values in *A. purpureocaeruleum*. Notably, we detected significant differences in LA even among populations of *A. calabrum*. However, the two species did not differ in their Grime's ecological strategies, showing the prevalence of stress-tolerant and ruderal components. Genetic diversity values were comparable between the two species ($H_e = 0.295$ in *A. calabrum* and $H_e = 0.311$ in *A. purpureocaeruleum*), indicating no evidence of genetic erosion in the endemic species. The comparison between genetic and phenotypic distances suggests that directional selection is the main evolutionary force shaping variation in both species. Nevertheless, two of the three *A. calabrum* populations exhibited niche conservatism, likely influenced by stabilizing selection. The result of this study challenges the widespread assumption that endemic species necessarily display low genetic variability and highlight the existence of different selective pressures among populations of narrow-ranged endemic species.

1) F. Selvi, G. Campetella, R. Canullo, S. Chelli, G. Domina, E. Farris, C. Gasperini, L. Rosati, C. Wellstein, E. Carrari (2023) The Italian endemic forest plants: an annotated inventory and synthesis of knowledge. *Plant Ecol. Evol.*, 156, 29–45

2) L. Cecchi, A. Coppi, H.H. Hilger, F. Selvi (2014) Non-monophyly of *Buglossoides* (Boraginaceae: Lithospermeae): Phylogenetic and morphological evidence for the expansion of *Glandora* and reappraisal of *Aegonychon*. *Taxon* 63: 1065–1078

1.8 = Population genetics, eDNA and ecological niche reconstruction as tools for the conservation of fragmented-range endemic plants: the case study of *Primula palinuri* (Primulaceae)

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Primula palinuri Petagna is a relict species endemic to southern Italy, with populations located along certain vertical cliffs of the Tyrrhenian coast over a stretch of approximately eighty kilometers. The peculiarity of the ecological niche occupied by *P. palinuri* defines it as the only Italian *Primula* adapted to live in coastal environments. To date, the species' range is highly fragmented, and the known populations consist of relatively few individuals, making this endemic species at risk of extinction.

The aim of this work is to develop a methodology useful for: (i) defining the conservation status of *P. palinuri* populations; (ii) identifying any critical issues related to inbreeding depression within the populations; (iii) identifying potentially suitable sites within the species' range for the establishment of new populations in order to overcome the limitations caused by the lack of gene flow.

Survey and sampling campaigns were carried out along the relevant coastal stretch. Known populations were confirmed, and one population not previously reported in the literature was found, bringing the total number of recognized populations to ten. In order to detect gene flow depression among populations, a population genetics analysis was conducted through the identification and quantification of Single Nucleotide Polymorphisms (SNPs). Once the necessary permits were obtained, plant material sampling was carried out using a strict protocol designed to minimize disturbance to leaf organs, collecting only the minimal portion of tissue required for DNA extraction. DNA was extracted and sequenced, SNPs were identified, and the key parameters for determining the degree of relatedness among populations were established.

During the collection of leaf material, environmental DNA (eDNA) present on inflorescences was sampled using sterile swabs, to detect genetic traces of arthropods and vertebrates and identify the main organisms involved in pollination. Physical and environmental parameters useful for defining the ecological niche were also recorded. The sampled populations consisted of an estimated 30 to 200 individuals each. Data obtained from the genomic analysis of the populations enabled the definition of population structure and the corresponding gene flow. Results showed that *P. palinuri* populations can be divided into distinct clusters, revealing a significant reduction in gene flow between these clusters. The eDNA analysis identified arthropods with limited flying capacity as the main pollinators, which are insufficient to ensure gene exchange between clusters. Due to physical separation caused by geographic barriers such as coastal morphology and the sea, and due to the distances between populations, inter-population variability is high, whereas intra-population variability is low. Characterisation of the ecological niche made it possible to propose, along the species' range, the reintroduction of populations—composed of individuals from existing clusters—into suitable intermediate sites. These would serve as potential stepping stones for pollinating insects. This could potentially restore gene flow interrupted by geographic barriers, allowing for increased genetic variability.

1.9 = Hotspots of alpine vascular plants of the Alps and their conservation status

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Mountains are biodiversity hotspots, and the proportion of endemics increases along the elevational gradient. Due to environmental conditions and historical processes, the distribution of vascular plants, and especially of endemics, is not uniform even within a single mountain range. In the Alps, the largest mountain range in Europe, the distribution patterns of endemics are strongly influenced by peripheral refugia during the last glacial maximum (1). Anthropogenic activities and ongoing climate change threaten alpine habitats and cold-adapted species (2). This creates a need for the protection of mountain areas. To make protection as effective as possible, it is useful to identify and protect hotspots of biodiversity. In this study, we used the IntraBioDiv database (3) which contains 991 subalpine and alpine species in the Alps, of which 345 are endemic. The hotspots were identified throughout the Alps using four grid-based biodiversity indices, i.e., species richness, endemic richness, weighted endemic richness, and weighted species richness. A grid cell was defined as a hotspot if it was within the top 5%, 5–10%, and 10–20% richest cells, respectively, for at least one of the indices. We considered a hotspot as a conservation gap, when less than 30% of its area size was within a protected area; this analysis was applied only to areas above 1600 m a.s.l.

Out of 388 grid cells, we identified 37 cells as top 5% hotspots, 32 cells as top 5–10 % hotspots and 60 cells as top 10–20% hotspots. Among those, 78,4%, 78,1% and 88,3% of the top 5%, top 5–10% and top 10–20% hotspots, respectively, were identified as conservation gaps. We strongly recommend the preservation of the identified conservation gaps, which are hotspots of alpine species with lack of adequate protection.

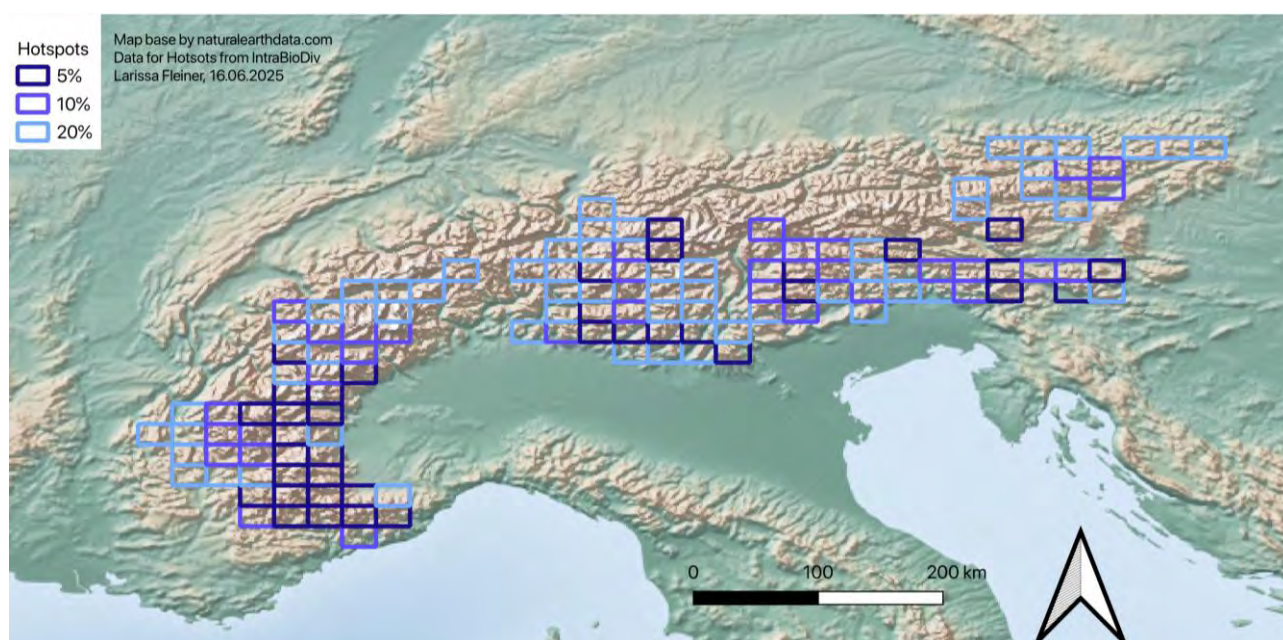


Fig. 1. The identified hotspots in the Alps, and the protected areas in this mountain range

- 1) P. Schönswetter, I. Stehlik, R. Holderegger, A. Tribsch (2005) Molecular evidence for glacial refugia of mountain plants in the European Alps, *Molecular Ecology* 14, 3547–3555
- 2) T. Dirnböck, F. Essl, W. Rabitsch (2011) Disproportional risk for habitat loss of high-altitude endemic species under climate change, *Global Change Biology* 17, 990–996
- 3) A. Tribsch, T. Englisch, F. Gugerli, R. Holderegger, H. Niklfeld, K. Steinmann, C. Thiel-Egenter, N. Zimmermann, P. Taberlet, Consortium I (2009) Integrating Data across Biodiversity Levels: The Project IntraBioDiv. In: Spehn EM, Körner C (eds) *Data Mining for Global Trends in Mountain Biodiversity*. Taylor and Francis, pp 89–105

1.9 = Biodiversity hotspots of alpine vascular plants in the Carpathian Mountain range and their conservation priorities

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Alpine ecosystems are highly sensitive to climate change, as rising temperatures expand warm-adapted habitats upward while shrinking cold-adapted alpine zones, a process particularly critical in vertically limited mountain systems (1). The Carpathians, one of Europe's most diverse mountain systems, host numerous endemic plant species and are increasingly threatened by both climate change and land-use (2).

In this study, we aim to identify alpine vascular plant diversity hotspots within the Carpathian Mountains and to assess their alignment with the existing network of protected areas. Using the IntraBioDiv dataset (3), the hotspots were quantified using three grid-based biodiversity indices, i.e. species richness, endemic richness, and weighted endemic richness. If a grid cell was within the top 5%, 5–10%, and 10–20% richest cells of the study area, respectively, for at least one of the indices, it was defined as a hotspot. When less than 30% of a hotspot area size was within a protected area, it was considered as a conservation gap. In total, 564 species have been recorded in the Carpathian Mountains, 59 of which can be categorized as endemic. There are 21 grid cells identified as top 5% hotspots, 16 grid cells are 5–10% hotspots, and 30 grid cells are 10–20% hotspots. These 67 hotspot grid cells will be spatially compared with current protected areas to detect conservation gaps - regions with high diversity but insufficient protection. This work will provide a baseline for prioritizing new conservation areas in the Carpathians and support evidence-based strategies for preserving alpine biodiversity under climate pressure.

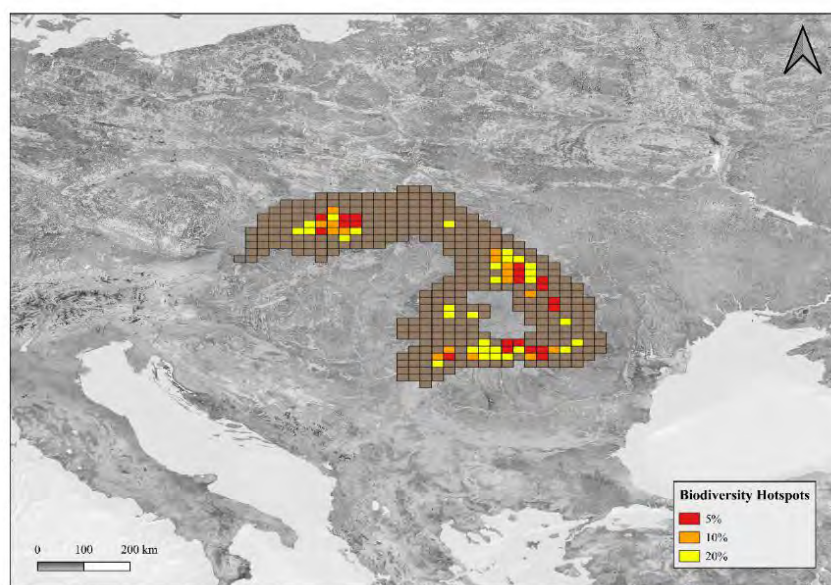


Fig. 1. Alpine plant biodiversity hotspots in the Carpathian Mountains. Red, orange and yellow grids are the top 5%, 5–10% and 10–20% hotspots

1) H. Essl, M. Staudinger, S. Dullinger, A. A. A. El Mokni, N. M. Mang, A. Milasowszky, J. Niklfeld (2009) *Biodivers. Conserv.*, 18, 567–582

2) T. Dirnböck, F. Essl, W. Rabitsch (2011) *Conserv. Biol.*, 25, 119–129

3) Tribsch, A., Englisch, T., Gugerli, F., Holderegger, R., Niklfeld, H., Steinmann, K., Thiel-Egenter, C., Zimmermann, N., Taberlet, P., Consortium I (2009) *Integrating Data across Biodiversity Levels: The Project IntraBioDiv*. In: Spehn EM, Körner C (eds) *Data Mining for Global Trends in Mountain Biodiversity*. Taylor and Francis, pp 89–105

1.9 = Evaluation of genetic and morphological diversity in *Tilia* spp in the campus of the University of Salerno (Italy)

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Habitat alteration resulting from persistent and intensive anthropogenic activities represents a major threat to the survival of plant and animal species, as well as to all living organisms more broadly. The wide range of human-induced pressures has increasingly drawn the attention of the scientific community, prompting efforts to develop innovative strategies for the conservation of the natural environment and the enhancement of biodiversity protection measures. The aim of our study was to evaluate the biodiversity within the *Tilia* trees present inside the Campus of the University of Salerno (Italy) through the combined use of molecular markers as the simple sequence repeats (SSRs) and the biometric analysis of informative morphological traits. The research involved 60 georeferenced *Tilia* trees, grouped into twenty Sampling Units (SUs) identified within the study area. Genetic characterization was carried out via DNA extraction followed by PCR amplification using six polymorphic nuclear microsatellite markers (Tc915, Tc31, Tc11, Tc6, Tc937, Tc7). The alleles detected at each SSR locus were encoded into a binary presence/absence matrix and subsequently subjected to numerical taxonomy analysis using dedicated software tools. A similarity dendrogram was constructed to evaluate the genetic similarity/diversity among the *Tilia* trees. The specimens analyzed were clustered into two main groups, one of which was further subdivided into two subgroups, revealing an overall similarity of 75% among all individuals. Morphological analyses were conducted based on specific morphological traits, including leaf margin and shape, fruit shape and size, apical bud size and color. The morphometric trait analysis showed the presence of three *Tilia* species and one hybrid, specifically, *T. cordata*, *T. platyphyllos*, *T. dasystyla* and *T. × europaea*. The patterns of genetic similarity revealed by means of molecular markers were consistent with the results obtained with the morphological traits. SSR analysis indicated a low level of polymorphism present in the *Tilia* trees studied. Despite the limited genetic diversity, notable differences were observed in the frequency of the species. Trees identified as *T. platyphyllos* and *T. cordata* were less frequent compared to *T. × europaea* and *T. dasystyla*. This concordance was further supported by the variations in lichen biodiversity observed on the bark of the trees. Kubiak and Osyczka (2020), Spier et al. (2010), well-documented the presence and frequency of epiphytic lichens can be influenced by bark pH and water-holding capacity. In the investigated area, which is a remarkable phenomenon for the “place” where it happened in, as in the case of SU2 and SU15, dominated by *T. dasystyla*, characterized by low lichen biodiversity. Conversely, a higher lichen biodiversity was recorded in those sites where *T. × europaea* hybrid was the prevalent one. This study contributes to the evaluation of genetic diversity among the *Tilia* individuals sampled within the UNISA Campus and underscores the importance of promoting plant species diversity in urban environments. Furthermore, it highlights the potential of selecting plant species that can serve as effective bioindicators for environmental monitoring.

2.1 = Characterization and future distribution prospects of the “Carciofo di Malegno” landrace (*Cynara cardunculus* subsp. *scolymus* (L.) Hayek) for its *in situ* conservation

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The loss of agrobiodiversity is a world damage both in biological and cultural aspects. Although mountainous areas in Italy still preserve many traditional cultivars (landraces), they face increasing risk from genetic erosion and the disappearance of landraces. They are valuable for their agronomic and phytochemical-nutritional properties and their ability to withstand climate change but only few traditional cultivars are scientifically characterized yet. The “Carciofo di Malegno” (figure 1) is a landrace of *Cynara cardunculus* subsp. *scolymus*, traditionally cultivated in the municipality of Malegno (Camonica Valley, Brescia Province, Lombardy, Northern Italy) already in the early 20th century and represents the only known artichoke landrace historically grown in the Italian Southern Alps. Local farmers describe “Carciofo di Malegno” as smaller, more bitter, and tastier than commercial varieties and its propagation is traditionally done by planting suckers in autumn or spring.

This study is aimed to characterize its morphological and phytochemical traits and to predict its future distribution prospects under climate change, supporting strategies for *in situ* conservation and sustainable enhancement. Morphometric analyses using geometric outline-based method revealed that the “Carciofo di Malegno” is morphologically distinct from commercial cultivars, displaying a spiky, conical shape similar to the “Spinosi” group and markedly different from the round-headed “Romaneschi” type. These results demonstrate the utility of geometric morphometrics as an efficient and objective method to support DUS testing in landrace characterization. Phytochemically, “Carciofo di Malegno” landrace exhibited a valuable profile comparable to commercial varieties, with significant levels of chlorogenic acid (497.2 ± 116.0 mg/100 g DW in heads; 1032.9 ± 294.9 mg/100 g DW in stems), cynarine (7.4 ± 1.2 mg/100 g DW) and luteolin (9.4 ± 1.5 mg/100 g DW) demonstrating high potential for food, health, and herbal applications. Species distribution modeling (MaxEnt, SSP2-4.5 scenario) indicated that climate change may shift the optimal growing area of this landrace to higher altitudes reaching a large part of the pre-Alps and Alps of Lombardy by the 2040–2060s. This trend highlights a paradox in current conservation regulations, which link landrace protection to traditional geographic areas, but it may promote the “Carciofo di Malegno” expansion, contributing to its cultivation and enhancement. The results underscore the value of this landrace as a genetic resource for breeding programs targeting cold-tolerant cultivars, and as a driver for sustainable agrobiodiversity-based development in mountain regions. Involving custodian farmers and adapting conservation frameworks to future climatic realities are essential to ensuring the long-term survival and valorisation of this unique genetic Alpine heritage.



Fig. 2. “Carciofo di Malegno” landrace

2.1 = Developing a Landscape Ecology-Based Diagnostic Methodology to guide Agrobiodiversity Conservation and Support in Peri-Urban Landscapes

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In the context of the ongoing decline in biodiversity within peri-urban landscapes (PUL) of the urban-rural interface, this study proposes a diagnostic tool derived from landscape ecology. The primary objective of this tool is to assist in the formulation of management strategies that are designed to preserve agrobiodiversity within these complex and ever-changing environments. A case study is presented related to Oasi Ca' Granda farm (South of the Metropolitan City of Milan), where there is an overlap between environmental degradation, landscape simplification and negative externalities related to land use. The method is divided into two primary stages (1). The initial step in the process is the “reading” of the landscape, which is achieved through a multiscale analysis of structural and functional elements. It considers diversity indices, patch structure and size, ecological connectivity and biotic providers (Fig. 1), allowing the understanding for both vulnerability drivers and ecological potential at both local and large territorial scales. The subsequent phase is the “writing” of the landscape, through which strategic interventions are cast by Nature Based Solutions (NBS). The purpose of such interventions is to stimulate ecological restoration processes, thereby supporting agrobiodiversity and improving the delivery of ecosystem services (2, 3). Strategic interventions are based on agroforestry practices and green and blue infrastructures. These are used as integrated tools to increase the structural and functional complexity of the PUL. These elements have the capacity to engender ecological resilience and facilitate biodiversity at various scales. The multi-scale study leads to the identification of a farm-scale masterplan, the intention of which is to serve as an evolutionary and replicable development framework. The combination of scientific instruments and local environmental scientific knowledge is a key aspect of the study. The outlined strategy has been shown to make the ecological contributions of management visible and accountable. It addresses ecological fragmentation, functional disconnections and the loss of spatial identity by providing a method to transform environmental pressures into an opportunity for systemic balancing and regeneration.

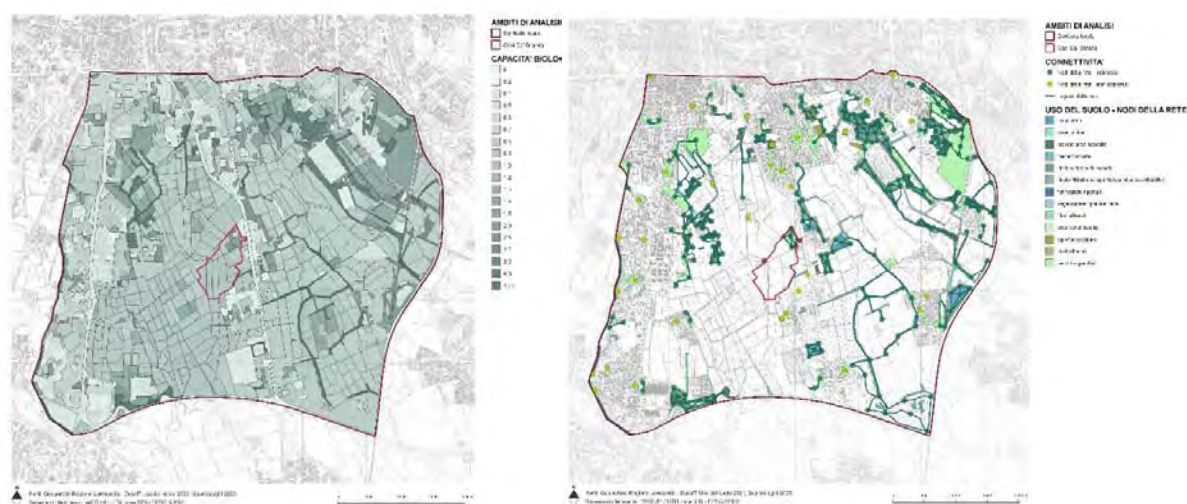


Fig. 1. Local landscape ecological diagnosis: Biological Territorial Capacity (BTC) map (left side); connectivity and circuitry graph analysis (right side)

- 1) Vagge, I., Chiaffarelli, G., Pirola L., Gibelli, M.G., Sgalippa, N., 2024. Landscape Ecology and Ecosystem services as Landscape Analysis and Assessment Tools for Ecological Landscape Planning. In: Dr. Sérgio L. (ed) Landscape Architecture and Design - Sustainability and Management. IntechOpen, Rijeka. <https://doi.org/10.5772/intechopen.1006355>
- 2) Vagge, I., Sgalippa, N., Chiaffarelli, G. 2024. Agricultural Landscapes: A Pattern -Process-Design Approach to Enhance Their Ecological Quality and Ecosystem Services through Agroforestry. Diversity, 16, 431. <https://doi.org/10.3390/d16070431>
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2.3 = Responses of Trees to Combined Drought and Heat Stress: Pressure–Volume Traits and Photosynthetic Performance

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Global temperatures have risen at an unprecedented rate in recent decades, increasing the risk of drought and heat stress on vegetation. These trends present a significant challenge to plant productivity and ecosystem stability. In this context, understanding the response of photosynthesis and plant water relations is crucial for identifying physiological traits associated with tolerance to environmental stress. We studied the physiological responses of six tree species to combined drought and high temperature: *Platanus acerifolia*, *Quercus robur*, *Quercus pubescens*, *Quercus petraea*, *Quercus pyrenaica* and *Quercus cerris*. The combined effects of water availability and temperature were assessed as follows: two sets of plants per species were subjected to either well-watered (*Relative Extractable Water*, 80%) or water-stressed (REW, 30%) conditions and maintained at an air temperature of 25 °C for three weeks. Subsequently, the same plants under both water regimes were subjected to an increased temperature of 30 °C and measured after 15 days under these conditions. The following photosynthetic parameters were measured: the photosynthetic assimilation rate under light and CO₂-saturated conditions (A_{MAX}), the CO₂ assimilation rate under light-saturated conditions at ambient CO₂ (A_N), the maximum rate of carboxylation by Rubisco (V_{cmax}), the maximum rate of electron transport (J_{max}), the triose phosphate utilisation (TPU), mesophyll conductance to CO₂ (g_m), stomatal conductance to H₂O (g_s), and intrinsic water use efficiency (WUE_i). Water relations traits were derived from pressure-volume curves: apoplastic water fraction (a_f), osmotic potential at full turgor and at the turgor loss point (π_o and π_{tlp} , respectively), leaf dry matter content (LDMC), relative water content at the turgor loss point (RWC_{tlp}), bulk modulus of elasticity (ϵ) and leaf capacitance (C). Under optimal conditions, π_{tlp} varied across species from -2.04 (*Q. cerris*) to -1.29 MPa (*P. acerifolia*), whereas all species showed a decrease under drought due to osmotic adjustment. High temperature did not affect the turgor loss point. However, some species showed minor elastic adjustments (decrease in ϵ) under elevated temperatures. No interaction between heat and water stress was observed in any water relation trait across species. Regarding physiological responses, the combined effect of high temperature and drought influenced net CO₂ assimilation (A_N). In *Q. cerris*, plants exposed to both high temperature and drought (HT-D) showed higher A_N values than those subjected to drought alone (CT-D). In contrast, *P. acerifolia*, maintained A_N at levels similar to those under drought-only conditions. Correlation analysis indicated that A_N was generally coordinated with stomatal conductance (g_s), with additional contributions from mesophyll conductance (g_m) and electron transport (J_{max}) under stress in some species. Regression analysis confirmed an increased sensitivity of A_N to g_s under drought in several *Quercus* species and treatment-dependent changes in the A_N - J_{max} relationship. Understanding the relationships among these physiological parameters can help identify functional traits that confer tolerance to water and thermal stress. This information will be essential for developing selection criteria for the most resilient species and for designing predictive tools to guide the sustainable management of urban and forest trees in the face of climate change.

2.3 = *Aspergillus sydowii*: An Emerging Threat to Mediterranean Corals from a Climate Change Perspective?

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Mediterranean Sea gorgonians, including *Eunicella cavolini* (Koch, 1887) (Fig.1) (1), play a crucial ecological role within coralligenous communities, acting as biodiversity hotspots (2). However, these species are increasingly vulnerable due to multiple stressors, including climate change and the emergence of opportunistic pathogens. This study was conducted in the Portofino marine protected area (Ligurian Sea) with the objective of analysing the fungal component associated with healthy and diseased *E. cavolini* fragments. The analyses conducted identified several fungal species, including *Aspergillus sydowii*, a well-known pathogen of Caribbean gorgonians (Fig.2) (3), reported here for the first time in Mediterranean corals. This prompts further inquiries into its potential role as a pathogen, with significant ramifications in a probable context of stress and decline for Mediterranean anthozoans. These results underscore the necessity for enhanced monitoring of the mycological component of marine ecosystems, and for investigation of the potential impact of emerging and non-emerging pathogens on the health of Mediterranean gorgonians.



Fig. 1 *Eunicella Cavolin*



Fig. 2 *Gorgonia ventalina* affected by *Aspergillus sydowii*

1) Sini, M., Kipson, S., Linares, C., Koutsoubas, D., & Garrabou, J. (2015). The yellow gorgonian *Eunicella cavolini*: demography and disturbance levels across the Mediterranean Sea. PloS one, 10(5), e0126253

3) Smith, G. W., Harvel, C. D., & Kim, K. (1998). Response of sea fans to infection with *Aspergillus* sp.(Fungi). Revista de biología tropical, 46(S5), 205-208

2) Trainito, E., & Baldaconi, R. (2016). Coralli del mediterraneo. Il Castello

2.3 = The core of the trees: where carbon reserves meet the dendrochronology

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The current climate crisis serious threat forest functioning, as tree migration potential and long-term genetic adaptation are too slow to cope with rapid changes in environmental conditions. Yet, we still have limited ability to predict climate-related tree decline and mortality, mostly because early-warning signals are often subtle and species-specific. To better predict the fate of forests, besides an in-depth understanding of how climate variables affect tree growth, it is crucial to investigate the relationship between anatomy and physiological mechanisms involved in tree acclimation responses. Indeed, dendrochronology is a powerful tool to assess eco-physiological drivers of tree growth at interannual resolution (1) and the potential to invest in an adequate pool of NSC should be taken into account (2).

Considering that trees archive their responses to stressors within their datable growth layers, we reconstructed xylem anatomical traits (i.e. ring width) (Fig. 1) and the ring-age-related distribution of non-structural carbohydrates (NSC) in trees of *Abies alba*. Six old trees (age > 50 years) were selected within a natural population of *Abies alba* and two core samples were collected for each tree. One core was used for dendrochronological measurements, in the other the NSC concentration was measured ring by ring. This could be used as a sensitive and effective diagnostic tool to predict which trees are more prone to suffer from extreme climate events, and in particular to drought-induced die-off.

Our data suggested that NSC concentration was correlated with annual growth variability and wood density. Moreover, we postulated that the ratio between soluble NSC and starch was modulated by water scarcity/availability, depicted by ring thickness.

We advanced the hypothesis that trees rely entirely on the remobilization of reserves stored during the previous growing season to support stem radial growth, yet this impacts the capacity of resistance to drought events. Future investigation calls for a thorough evaluation of different *Abies alba* populations for a rigorous evaluation of the link among genotypes, phenotypes, physiological traits, and climatic drivers constraining tree growth across time and environments.

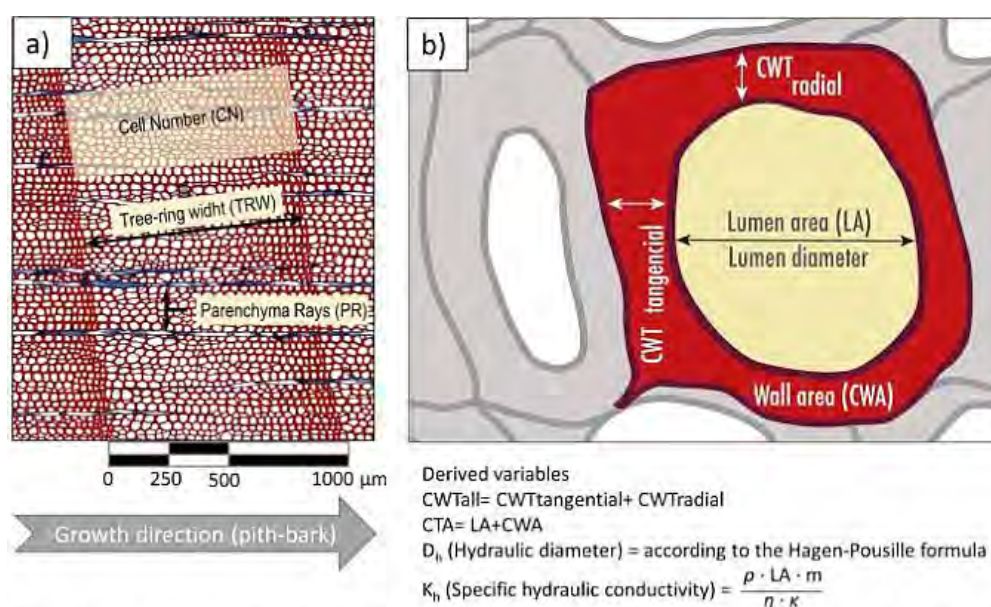


Fig. 1a-1b. Conifer tree ring

1) D. Frigo, P. Römer, L. Unterholzner, H. Zimmer-Zachmann, J. Esper, M. Carrer, E. Ziacco (2024) Dendrochronologia, 88: 126241

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2.3 = Mediterranean forest vulnerability to climate pressure: the case of *Quercus ilex* L. dieback and mortality in central and southern Italy

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Forest vulnerability to climate change has significantly increased in recent decades, affecting the vegetation productivity and health. Climate-driven hotter droughts are leading to widespread canopy dieback episodes, rising tree mortality rates and today representing a significant threat to forests, also in Mediterranean areas, where evergreen sclerophyllous vegetation is considered resistant and adapted to drought stress.

In Italy, the first reports of dieback of *Q. ilex* forests due to climatic events date back to 2012 and, in a more extensive manner, concern the extreme drought in the year of 2017 (1, 2). During the summer of 2024 a widespread decline occurred in several regions of central (Tuscany) and southern Italy (Apulia and Basilicata) and on the islands (Sicily and Sardinia, fig. 1a, 1b). Symptoms of dieback, such as total or partial leaf browning (fig. 2a, 2b, leaves and twigs desiccation, emission of epicormic shoots, crown defoliation and tree mortality were observed on evergreen sclerophyllous vegetation, mainly in *Quercus ilex* L. and *Q. suber* L. stands and shrubs (*Myrtus communis* L., *Phillyrea* sp., *Arbutus unedo* L., *Pistacia lentiscus* L.). The decline events were often associated with pathogens, such as *Phytophthora* species and *Biscogniauxia mediterranea*. This contribution provides an overview of the current Mediterranean evergreen vegetation decline in Italy, which is now of national importance. Due to the severity and increasing frequency of these decline events, there is an urgent need to improve forest monitoring networks, with regional initiatives aimed at assessing the dynamics of the environmental drivers (climate, site, management) and impacts, by means of a multidisciplinary approach, with ecological, physiological and phytopathological investigations, as well as the involvement of forest managers and citizens in the early detection of impacts.



Fig. 1a-1b. *Quercus ilex* dieback in Sardegna (Ogliastra, August 2024) and details of dried trees



Fig. 2a-2b. Leaf symptoms (desiccation and browning development in *Quercus ilex*)

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2.4 = How to Save a Botanical Paradise: Implementing REDD+ in Papua New Guinea

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Papua New Guinea (PNG) harbors one of the world's most significant tropical forest ecosystems. Approximately 80% of the country's land area, over 38 million hectares, is classified as forested, comprising the third-largest tropical forest area globally, following the Amazon and Congo Basin. Of this, more than 75% (circa 28 million hectares) remains 'intact,' exhibiting minimal disturbance from anthropogenic activities. PNG is a global biodiversity hotspot, supporting over 5% of the planet's known biodiversity. The country's flora is particularly diverse, with 13,634 documented vascular plant species and estimates ranging from 15,000 to 20,000 species, representing the richest island flora worldwide (1). Despite its ecological importance, PNG's forests are experiencing significant deforestation, particularly in lowland areas, which constitute approximately 20 million hectares. Since 2010, forest loss in these regions has proceeded at an annual rate of 3–5%, predominantly driven (>90%) by industrial logging for export. In support of sustainable forest management and the implementation of Reducing Emissions from Deforestation and Forest Degradation (REDD+) and other conservation finance mechanisms, a multipurpose National Forest Inventory was conducted. The inventory aimed to generate baseline data on carbon stocks and biodiversity, focusing on both floristic and faunistic components (2). Field data were collected from 117 circular plots, distributed across an altitudinal gradient and including unlogged and logged forest sites. The study examined the influence of altitudinal gradient and logging disturbance on several taxonomic groups: trees (Fig. 1), ferns (Fig. 2), lianas, herbaceous plants (Fig. 3), ants, and moths. Results revealed differential patterns in species richness, diversity, and composition among these groups in response to elevation and logging activities. Beside a significant improvement of the taxonomic knowledge of an understudied area, this research provided a foundational ecological baseline to support long-term monitoring of forest condition and biodiversity within the context of REDD+ implementation. The findings underscore the importance of conserving PNG's forest ecosystems, which offer substantial ecological, economic, and social co-benefits.



Fig. 1. *Syzygium malaccense* (L.) Merr.



Fig. 2. *Dipteris conjugata* Teinw



Fig. 3. *Cyrtandra hololeuca*
B.L.Burt. & L.M. Perry

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2.4 = Preliminary assessment of *Tuber melanosporum* mycorrhization in a long-unmanaged truffle orchard

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This study aims to investigate the evolution of mycorrhization in a *Tuber melanosporum* truffle orchard that has not been managed for over 19 years (Fig. 1). The research focuses on the Banditelle I experimental truffle ground, established in 1983 as part of the Regional Truffle Program (PTR) promoted by the Umbria Region. This program led to the creation of 59 experimental and demonstrative truffle orchards across the region, covering approximately 115 hectares and planted with symbiotic trees inoculated with valuable truffle species. Between 1997 and 2009, various monitoring and experimental activities were carried out to assess the status of the orchards, the degree of mycorrhization, and the effectiveness of different cultivation techniques. Banditelle 1 (Fig. 2), was selected as one of nine sites for agronomic trials. These agronomic trials yielded particularly interesting data regarding the percentage of trees mycorrhized with *T. melanosporum* and other ectomycorrhizal fungi (1). Scientific literature highlights that the persistence of *T. melanosporum* mycorrhiza is influenced by agronomic management (2). In particular, the absence of cultivation practices can promote the decline of specific symbioses in favor of generalist fungi or opportunistic competitors (3). Within this context, it is hypothesized that the percentage of trees mycorrhized with *T. melanosporum* has significantly decreased due to interspecific competition and the natural aging of the plantation. The study is currently ongoing. Planned activities include sampling and analyzing the roots of a representative number of trees within the orchard, with the goal of assessing the presence and identity of symbiotic fungi. The interpretation of the data, integrated with available historical information, will help provide a comprehensive understanding of mycorrhizal dynamics in unmanaged systems, offering insights for the conservation and potential recovery of truffle plantations. This study was supported by: “Monitoring and management of natural and cultivated truffle areas in state-owned territory” AFOR local project; INTACT RISE-MSCA Project, Grant Agreement n. 101007623.



Fig. 1. Experimental truffle ground in 2024



Fig. 2. Experimental truffle ground in 2004

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2.4 = Post-fire vegetation dynamics in Mediterranean pine plantations under naturalistic management with burnt logs *in situ*

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Fire is a major ecological driver in shaping Mediterranean landscapes, exerting significant influence on ecosystem structure and composition through processes such as fragmentation, transformation of natural systems, and the formation of secondary habitats (e.g., garrigue and steppe grasslands) (1). These dynamics lead to increased environmental heterogeneity and biodiversity at the landscape scale. Post-fire regeneration is governed by multiple abiotic and biotic factors, including fire intensity and type, as well as prevailing climatic conditions. Plant species exhibit diverse fire-adaptive traits; however, the introduction of alien species can facilitate biological invasions, potentially undermining the resilience and recovery of native plant communities (2). Pine forests represent one of the most fire-prone vegetation types in the Mediterranean basin, making the study of post-fire successional dynamics particularly relevant for forest management and conservation strategies. In this context, a field study was conducted in pine stands afforestation with *Pinus pinaster* subsp. *pinaster* and *Pinus radiata* affected by a wildfire in 2021, located in the municipality of Roccaforte del Greco (Metropolitan City of Reggio Calabria, Southern Italy), at elevations ranging from 970 to 1,020 meters above sea level, partly affected by Nature-Based arrangements using felled burnt logs by wildfires as anti-erosion barriers (3). Vegetation surveys conducted in the years 2024-2025 with the transect method highlight the onset of auto-successional processes within the pine forest area destroyed by fire. Notably, a significant expansion of herbaceous species typical of open and disturbed environments and pasture habitats was recorded, promoted by the remarkable reduction in tree canopy cover. Among the most represented taxa there were *Pteridium aquilinum*, *Dactylis glomerata*, *Sinapis pubescens*, *Achillea ligustica*, *Thapsia garganica*, *Hypochaeris radicata*, *Euphorbia rigida*, and *Crepis vesicaria*, alongside synanthropic and ruderal species such as *Vicia sativa*, *Medicago hispida*, *Ornithopus compressus*, *Sonchus asper*, *Chenopodium album*, and *Geranium rotundifolium*. The floristic composition also included some alien species: *Erigeron bonariensis* and *Cupressus sempervirens*, whose presence—although currently limited—may represent a potential factor in altering future successional trajectories. Simultaneously, the reappearance of pre-existing nemoral species such as *Pimpinella anisoides*, *Geranium robertianum*, *Poa sylvicola* and *Cyclamen hederifolium* was documented, suggesting a partial preservation of the original floristic pool. The recolonization by woody species previously established in the area was evident at both the shrub layer (*Spartium junceum*, *Rosa canina* - growing in xeric, south-exposed slopes - *Cytisus villosus*, and *Cytisus scoparius* - in mesic, north-exposed slopes) and the tree layer, composed of the same species that originally formed the plantation (*Pinus radiata*, an allochthonous species and *Pinus pinaster* subsp. *pinaster*). The latter, however, do not represent the potential natural vegetation of the site, as they originated from past afforestation interventions and were subsequently affected by the fire. The potential natural vegetation of the area is instead represented by woodlands dominated by *Quercus pubescens*, as evidenced by the presence of oak tree clusters in the surrounding areas. The sporadic presence of *Q. pubescens* regeneration highlights the potential of this species to gradually recolonise the site through secondary succession, provided that no further disturbance events occur. In conclusion, the post-fire vegetation dynamics observed in the pine forests of Roccaforte del Greco demonstrate the initiation of natural successional processes characterized by a shift from fire-adapted herbaceous and ruderal species toward the gradual re-establishment of shrub vegetation. While current recolonisation also includes species from past artificial plantations, the eventual recovery trajectory is expected to favor the native climax community dominated by *Q. pubescens*, contingent upon the absence of further disturbances. The presence of alien species, albeit limited at present, underscores the need for ongoing monitoring to mitigate potential impacts on native biodiversity and successional pathways. These findings contribute valuable insights into Mediterranean forest resilience and inform adaptive management strategies aimed at conserving ecosystem integrity in fire-prone landscapes.

This study was carried out within the project “TECH4YOU – Technologies for climate change adaptation and quality of life improvement” and received funding from the European Union Next-GenerationEU (National Recovery and Resilience Plan (PNRR) - M4C2 - Investment 1.5 – “Innovation Ecosystems” - D.D. 3277 of 30 December 2021).

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2.4 = Soil microbiome dynamics in white truffle habitat of the Motovun forest (Croatia): Insights from a Mediterranean floodplain ecosystem

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The Motovun forest (Fig. 1), located in the Mirna River valley on the Istrian Peninsula (Croatia), is one of the last lowland Mediterranean alluvial forests and a key natural habitat for the prized white truffle (*Tuber magnatum* Picco). This study investigates the soil microbiome and vegetation of this unique ecosystem to understand the ecological conditions in different forest areas and their relationship to the characteristics suitable for the presence of white truffles. The analysis was conducted across four designated areas, as determined from prior research (1). Within these areas, three specific sub-areas were selected for detailed vegetation analysis and soil sampling. Soil samples were collected at two depths (15 cm and 30 cm) during the months of July and November. These samples were used for chemical-physical evaluations and, following the extraction of environmental DNA, for a spatio-temporal characterization of the bacterial and fungal communities in the studied regions. To characterize these communities, high-throughput amplicon sequencing was employed, emphasizing their diversity, composition, and potential interactions between different taxa and their environment. Soils that support the growth of white truffles, which typically experience sediment recharge, contain distinct biological communities compared to areas that are seasonally saturated or extremely dry. We identified several Operational Taxonomic Units (OTUs) commonly associated with *Tuber magnatum*, even in the absence of visible fruiting bodies. This suggests the potential presence of truffles and indicates a strong environmental filtering effect. This research highlights the ecological complexity of the Motovun Forest, where anthropogenic changes, such as historical mismanagement of the forest and recent hydrological alterations, have affected soil conditions and, consequently, the microbial ecosystem. As more than 300 hectares of this forest are now protected within the Natura 2000 network, understanding its microbiome offers crucial insights into sustainable forest and truffle management. These findings contribute to the broader field of soil microbial ecology in Mediterranean floodplain forests and support conservation efforts related to both biodiversity and rural truffle economies.



Fig. 1. The Motovun forest (Croatia)

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2.6 = *Quercus cerris* L. fine root adaptation across an urbanization gradient

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Urbanization has expanded rapidly worldwide and is projected to increase further in the coming decades (1). This global trend profoundly affects ecological processes in urban ecosystems, leading to biodiversity loss, carbon stock depletion, habitat fragmentation, and various forms of environmental pollution: air, water, light, and noise along with altered microclimatic conditions such as the urban heat island effect (2). In this context, urban forests (UFs) play a critical role in mitigating the negative impacts of urbanization and enhancing both environmental quality and human well-being. UFs provide essential ecosystem services, including biodiversity conservation, soil erosion control, stormwater regulation, pollutant absorption, air filtration, CO₂ sequestration, and climate change mitigation (3). However, the functionality and benefits of urban forests are strongly dependent on their ecological integrity, which is often compromised by anthropogenic pressures. Urban forests are highly susceptible to fragmentation and degradation, resulting in heterogeneous and frequently disturbed soil environments that ultimately affect plant performance and forest resilience. In this complex and dynamic context, plant functionality, assessed through functional traits, offers a valuable lens for understanding ecosystem responses to urban stressors. Functional traits such as nutrient acquisition strategies, stress tolerance mechanisms, and contributions to biodiversity allow researchers to evaluate how plant communities and individual species adapt to and shape their environments. The rhizosphere in urban environments plays a key role in plant health and ecosystem functioning, yet its spatial variability remains poorly understood, particularly in forest trees. This study investigates the variation of fine root functional traits of *Quercus cerris* L. across topsoil and subsoil layers in non-urban, peri-urban, and urban forests, with the aim of understanding how root traits respond to urbanization. We have selected three sites characterized by increasing levels of fragmentation and decreasing canopy cover. Seven *Quercus cerris* L. trees with similar stem diameters were selected for each site. Each tree, located within approximately 30-50 meters of its nearest neighbor, was considered an independent replicate. Trees were sampled in spring. Two 30 cm-deep soil samples were collected from each tree using a soil core sampler, divided into 10 cm segments, and processed to extract fine roots (≤ 2 mm in diameter). Fine roots were classified into two groups: *Q. cerris* and other species. Roots were identified based on their morphology and anatomy, with oak fine roots further separated into live (biomass) and dead (necromass). The roots were weighed and scanned and analyzed for length by diameter class. Fine root specific root length (SRL) was calculated to assess root dynamics. In non-urban forests, fine roots were predominantly shallow, and their turnover contributed to carbon cycling through the accumulation of root-derived necromass in the topsoil. In peri-urban areas, fine root traits exhibited marked vertical differentiation, with higher specific root length in the top layer, potentially linked to microhabitat fragmentation. In urban forests, root interactions shifted deeper into the soil profile, affecting root length and biomass, while overall trait variability decreased—suggesting environmental disturbance or functional constraint. We hypothesize that these spatial changes in fine root traits reflect altered resource acquisition strategies and could influence ecosystem processes such as nutrient uptake and organic matter turnover. The study emphasizes the importance of assessing belowground plant traits to better understand how urbanization alters forest functioning. These findings highlight the adaptive role of fine roots in responding to heterogeneous and anthropogenically influenced soil environments.



Fig. 1a-1b. Sampling of soil cores

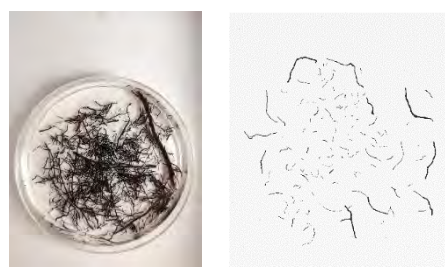


Fig. 2a-2b. *Quercus cerris* fine roots

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2.7 = The inner membranes of the pomegranate (*Punica granatum*) fruit have protective properties on the human neural line SH-SY5Y

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The pomegranate (*Punica Granatum*, L.) is a plant of the genus *Punica*, belonging to the family Lythraceae (previously Punicaceae) [1]. *P. Granatum* is native to northern India, but it has been widely cultivated in Central Asia, China, and throughout the Mediterranean region [2]. *P. Granatum* is a small deciduous tree or shrub with a bushy growth habit; its fruits are berries, and the diameter ranging from 5 to 12 cm. They have a smooth, coriaceous, reddish-brown outer shell. The inner part of the fruit is divided into lodges delimited by a yellow, spongy tissue, which is not edible. The lodges contain numerous "grains" of prismatic shape, contiguous and edible. Each grain consists of a translucent, fragrant, and juicy pulp, garnet-red color, surrounding a small woody seed. Pomegranate demonstrates numerous protective properties on human health (anti-inflammatory, antioxidant, anti-cancer, antibacterial, and antiviral) justified by the countless chemical components that include amino acids, minerals, vitamins, polyphenols, flavonoids, alkaloids, steroids, phenylpropanoid compounds, triterpenes (volatile oils), fibers [3], and organic acids. The plant is represented in Figure 1.

We experimentally used and compared two extracts from the fruit:

1) The extract from the whole fruit (including the peel) that was pulverized (TOT-E).
2) The extract from the inner membranes of the fruit (which is not edible) is removed and pulverized (IM-E).
The two extracts have demonstrated antioxidant and anti-inflammatory properties on a cell line of human neurons (SH-SY5Y), but IM-E was significantly more effective than TOT-E. The amounts of polyphenols and flavonoids present in the two extracts were evaluated, and they were similar. In contrast, the amount of fiber in IM-E was statistically higher than in TOT-E. It would be interesting to see if the fiber is responsible for the activities found and if its removal cancels these properties.



Figure 1. Botanical representation of the sections of *P. Granatum*. Each box is marked with an alphabetical letter, corresponding to a district of *P. Granatum*. A = Bushy bearing of the plant; B = *P. Granatum* shrub with flowers in evidence; C = contorted trunk; D = lanceolate leaves; E, F = red-orange flowers; G = wilted flower; H = development of the fruit; I = branch with whole fruits; L = damaged ripe fruit, attached to the plant; M = open fruit with prominent grains; N = seeds contained in the grains. The images in Figure 1 were obtained and modified by www.actaplantarum.org/flora/flora_info.php?id=6272.

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2.7 = Towards the certification of urban forest ecosystem services through innovative tools and methodologies: the case of Ruffini Park in Turin

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Urban green areas play a crucial role in enhancing the quality of life in cities by providing numerous ecosystem services, including climate regulation, carbon sequestration, air pollution reduction, and the psychophysical well-being of the population. However, for these benefits to be properly recognized and valued, a methodology for quantification and certification based on reliable scientific tools is essential.

In this context, PEFC (Programme for the Endorsement of Forest Certification) aims to extend its approach to urban green spaces by introducing criteria for sustainable management and the estimation of ecosystem services.

This research aligns with that vision and aims to develop and test an innovative methodology for quantifying ecosystem services in urban areas, with a particular focus on Ruffini Park in Turin. The approach combines technologically advanced tools, such as LiDAR (Light Detection and Ranging) and GNSS (Global Navigation Satellite System), with data analysis software including LiDAR360 and i-Tree Eco.

The research activities were organized into several phases. First, a survey campaign was carried out using a laser scanner and GNSS antenna to obtain a high-resolution georeferenced point cloud, from which biometric information was extracted for a representative sample of trees in the park. The collected data were processed using Faro Connect and LiDAR360, enabling the determination of key parameters such as total tree height, crown diameter, and trunk diameter. In parallel, manual surveys were conducted to gather visual data on the trees, essential for integrating the input data required by the i-Tree Eco software.

Subsequently, i-Tree Eco was used to provide a detailed estimate of the ecosystem services offered by the green area, including the removal of air pollutants (NO₂, SO₂, O₃, CO, PM₁₀, PM_{2.5}), carbon sequestration and storage, avoided stormwater runoff, oxygen production, and the economic valuation of these benefits. The results highlighted the essential role of Ruffini Park in delivering ecosystem services to the city of Turin, confirming the importance of targeted management strategies to maximize benefits and minimize potential negative impacts.

The integration of advanced technologies, such as LiDAR, with simulation models allows for greater accuracy in quantifying ecosystem services and supports the implementation of data-driven management strategies. The application of this methodology could serve as a replicable model for other urban green spaces, contributing to the enhancement of PEFC certification standards in urban contexts. Looking ahead, these approaches could support public administrations and urban green space managers in planning more sustainable and effective interventions, promoting sustainability-oriented land management and improving the quality of life for citizens.

2.8 = The enigma of *Laccaria laccata* (Hydnangiaceae, Basidiomycetes)

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Laccaria laccata (Scop.) Cooke, the type species of the genus *Laccaria*, has long been recognized as one of the most widely distributed fungal species in the Northern Hemisphere (1). A simple Google search for the binomial name *Laccaria laccata* yields approximately 160,000 results, while Google Scholar indicates 7,230 citations, and Scopus indexes 291 papers concerning this taxon. These metrics demonstrate the significant attention that *L. laccata* has garnered from both scientific communities and the general public. Despite its reputation as one of the most easily identifiable species within its genus, molecular phylogenetic analyses have revealed a high degree of genetic diversity among specimens named as *L. laccata*, with sequences organizing into nearly 30 distinct terminal taxa or clades. This genetic complexity presents challenges to traditional morphological classifications and emphasizes the necessity for a reassessment of the species' taxonomy. Our research involved a comprehensive morphological and molecular analysis (using nrITS, nrLSU, and *RPB2* regions) of the specimen that Singer designated as lectotype of *L. laccata*, collected in Femsjö, Sweden, a location where Elias Magnus Fries conducted his pivotal studies. Furthermore, we collected and analyzed recent specimens of *L. laccata* in Femsjö region and other localities in Italy and England. This study also critically evaluates the typification process proposed by Singer, presenting an updated perspective on the species' classification based on modern methodologies.

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2.8 = A neglected forest species endemic to Brescia and Garda Prealps? Insights into the systematics of *Cardamine baldensis* (Brassicaceae)

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The western European *Cardamine heptaphylla* (Vill.) O.E.Schulz and the south-western European *C. pentaphyllos* (L.) Crantz are two forest species widely distributed across northern Italy, with largely overlapping ranges and occasionally co-occurrence at the same sites. Both species share the same chromosome number ($2n = 48$) (1), and are morphologically distinct: the former has usually white flowers, 7–9-imparipinnate basal and lower cauline leaves with a glaucous-green abaxial surface, and rhizomes bearing short scales (1–1.5 mm); the latter bears usually violet flowers, 5-palmate basal and lower cauline leaves green on the abaxial surface, and rhizomes with long scales (6–8 mm) (2). A sterile hybrid, *C. ×digenea* (Gremli) O.E.Schulz, morphologically intermediate between the two, has been sporadically recorded from several sites across southeastern France and Switzerland (2). These hybrids display rose-colored flowers, 5–9-imparipinnate to digitate basal and lower cauline leaves with an abaxial surface ranging from glaucescent to green, and rhizomes bearing both short and long scales. Later, Otto W. Sonder described a new taxon from the southwestern Garda Prealps (3), *Dentaria intermedia* Sond., which is morphologically similar to the hybrid but differs substantially in key traits, including full fertility, distinct leaf coloration, and rhizome characteristics. Monographer Otto E. Schulz, primarily based on the shared leaf and rhizome features, reclassified this unit of diversity as a variety of *D. pinnata* Lam. (= *C. heptaphylla*), a view that has persisted in most subsequent taxonomic treatments. However, due to its distinctive morphology and geographically restricted distribution, Karl Fritsch considered this taxon a separate species, renaming it *C. baldensis* Fritsch ($\equiv$ *D. intermedia* Sond., non *C. intermedia* Steud.). To clarify the taxonomic status of these populations in relation to both *C. heptaphylla* and *C. pentaphyllos*, we started a systematic study integrating field observations with nomenclatural, morphometric, cytogenetic, and molecular phylogenetic data. To this end, original material or nomenclatural types of the three taxa were located and examined. We sampled a total of 90 individuals from nine populations in northern Italy: three attributed to *C. baldensis* (two in the type locality, Tremalzo and Pur, and one from Monte Baldo); three to *C. pentaphyllos* (two in Monte Baldo and one near Lago d'Iseo); three to *C. heptaphylla* (one near Lago d'Iseo and two in the northern Apennines). We measured 27 morphological characters (22 quantitative and 5 qualitative), and collected leaf fragments preserved in silica gel and rhizomes for further analyses. Additionally, we closely examined photographic records available on iNaturalist (<https://www.inaturalist.org>) to assess the geographic distribution of *C. baldensis*. Our results suggest that this latter species is morphologically intermediate between *C. heptaphylla* and *C. pentaphyllos*, yet it shows a stronger overall resemblance to the former. This is supported by *a priori* classification (LDA) based solely on quantitative data, which correctly identified 94.4% of specimens (jackknifed), with only a few misidentifications between *C. baldensis* and *C. heptaphylla*. Despite this, *C. baldensis* can still be clearly distinguished by a combination of qualitative morphological characters that, together with the quantitative ones, allow a safe identification. Notable diagnostic features include its white-rose to violet flower coloration and the occurrence of at least some 5–7-digitate leaves. The two species are allopatric: the eastern distribution limit of *C. heptaphylla* corresponds to the mountains bordering the eastern shores of Lago d'Iseo, whereas *C. baldensis* is distributed from the contiguous Val Trompia eastwards to Monte Baldo. Taken together, morphological and geographical evidences strongly support the recognition of *C. baldensis* as a distinct species, representing another notable example of an Italian forest endemic. To determine whether *C. baldensis* originated from a hybridization event between two congeneric species or diverged allopatrically from *C. heptaphylla*, cytogenetic data (genome size) and molecular phylogenetic data based on ITS sequences are currently being collected.

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2.9 = The soil seed bank of managed and abandoned *Quercus cerris* coppice forests: evidence from an experimental study in Tuscany

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The soil seed bank consists of the collection of spermatophyte seeds that are incorporated in the soil of a given plant community or ecosystem. These seeds can persist and remain viable for a variable, and often unknown, number of years—ranging from one year to several decades. In forest ecosystems, they play a key role in triggering vegetation recovery after canopy disturbances such as wildfires or logging. The seed bank also represents a reservoir of hidden or “potential” plant diversity, often differing from the “realised” plant community in terms of species richness and composition (1). Despite its ecological importance, the diversity, composition, and ecological-functional profile of the soil seed bank remain poorly understood, especially in Mediterranean forest ecosystems (2). Even less is known about the legacy effects of long-term anthropogenic disturbances. These include intensive forest management practices, for example, coppicing.

To address these knowledge gaps, we investigated the soil seed bank of a supramediterranean Turkey oak (*Quercus cerris* L.) woodland located within the Berignone-Tatti forest (Volterra, Tuscany) under two different managements conditions: 1) active coppice-with-standards (last harvested eight years ago), 2) abandoned coppice-with-standards (unmanaged for about seventy years). Seed bank density, diversity and composition were assessed using a nine months’ experiment of controlled germination in the greenhouse. For each management type, 1.5 l of soil were collected in winter from four 5 × 5 m quadrats, grouped into three plots. Soil samples were placed in boxes under standardized conditions of temperature, light, and moisture to stimulate the germination of all viable seeds. Concurrently, a complete vegetation survey of the 24 quadrats was carried out to characterize species diversity, composition and structure of the “realised” plant community. Two complementary approaches were then used to analyse differences between the two managements on seed bank dynamics: i) a *horizontal* comparison between the seed banks of the managed and the abandoned coppice stands; ii) a *vertical* comparison between the seed bank and the realised vegetation within each management type.

Regarding the horizontal comparison, a higher species diversity was observed in the active coppice, with a marked compositional divergence between the two management types. Many species were annuals with generalist or ruderal tendencies, and several were non-native. *Erigeron canadensis* L. emerged as the most frequent species in both the active and the abandoned coppice. Other frequent taxa included *Cardamine hirsuta* L., *Eupatorium cannabinum* L., and *Juncus bufonius* L. Forest specialists were generally rare, but their proportion increase in the abandoned coppice. In the vertical comparison, an inverse relationship in species richness was detected between the seed bank and the realised plant community of the understorey in the two management types. In the active coppice, the realised plant community was richer than the seed bank (79 vs. 60 species), whereas in the abandoned coppice, the seed bank hosted a significantly richer potential flora than the realised vegetation (39 vs. 16 species). This suggests that numerous species persist in the seed bank, remaining dormant until significant and sudden disturbances makes microclimatic and light conditions more favourable for germination, such as forest harvesting. Overall, floristic divergence between the two forest types was greater in the realised plant community than in the seed bank, indicating that many forest species remain in the soil as a shared potential species pool. This study points to the impact of coppice management on the soil seed bank in thermophilous oak woodlands, mainly consisting in the increase of species richness but also in the ruderalization of its floristic composition.

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2.9 = Integrating the NBFC BEF monitoring network with old-growth forest sites: the case study of Sas Baddes holm oak forest in Sardinia

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Within the National Biodiversity Future Center Spoke 04, the project activity no. 4.1 focuses on advanced systems for monitoring, studying, and managing biodiversity, its organization, and the related ecosystem functions and services in relation to natural and anthropic impacts. With this aim, a network of selected areas was established to monitor biodiversity and its relationship with ecosystem functions. In this framework a selection of forest sites that are potential candidates for the designation as *boschi vetusti* (i.e., old-growth forests) will be included in the national network and investigated with a cross-taxon methodology. Following the National Decree of 5 April 2023, which establishes the Italian network of old-growth forests, the Sardinian Forest Corps (CFVA) has requested UNISS to identify the first set of three old-growth forests to set up the methodology for Sardinia.

Here we report the preliminary results of the cross-taxon investigation at Sas Baddes holm oak forest (*Aceri monspessulani-Quercetum ilicis*). The Sas Baddes forest, located in the Supramonte - Gennargentu forest complex, is renowned for its natural, aesthetic, and landscape value, having been the focus of several landmark ecological studies. Forest measurements (dendrometric survey) required to evaluate the criteria to meet for an old-growth forests (as in the Ministerial Decree of the 18th November 2021, GU no. 303 of the 22nd December 2021) were assessed in a 1-hectare circular plot, resulting in a total basal area slightly higher than 67 m²/ha. DBH, height and dendro-micro-habitat were recorded for all trees within the plot. A remarkable presence of standing deadwood and lying deadwood (CWD and FWD, with about 96 m³/ha of CWD) was observed. The occurrence of seedlings was evaluated using a random transect by counting all plants below the height of 10 cm in the 50 cm on either side of the transect. Information on vertebrates was provided by FoReSTAS on a historical basis, while Soil Biological Quality index (QBS-ar) was assessed during 2025. The investigation on the site included the survey of deadwood insects, Fungi, Lichens and Bryophytes. Furthermore, a paired sampling of eDNA is planned in the old-growth forest and in a nearby control area to get additional information on the soil biodiversity, as well as a LiDAR acquisition by UAV.

The present case study exemplifies how integrating the NBFC BEF monitoring network with old-growth forest ecosystems can provide valuable insights into biodiversity assessment and forest conservation across Italian forest types.

3.1 = A Novel Split-Root Hydroponic System for Investigating Inter-Plant Responses to Abiotic Stress Signals

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Traditional experimental approaches for studying inter-plant communication often rely on compartmentalising root systems, which can inadvertently alter natural physiological interactions. To address these limitations, we developed an innovative hydroponic system featuring a dual-compartment spatial design, consisting of a Free Sector (FS) and a Confined Sector (CS). This custom 3D-printed architecture guides unidirectional root growth from the FS to the CS through curved, non-linear channels embedded in the central septum. These channels are engineered to allow root passage from FS to CS while preventing root intrusion in the opposite direction. The system incorporates a precisely regulated drip irrigation setup and a high-percolation substrate (Fig. 1), enabling the targeted induction of water stress (via 20% polyethylene glycol, $\psi_w = -0.5$ MPa) in individual plants. Importantly, this design maintains belowground continuity between co-cultivated conspecific or heterospecific individuals, preserving natural root-mediated communication. Two model species - *Solanum lycopersicum* L. and *Tagetes patula* L. - were selected for co-cultivation experiments. After approximately one month of growth, once root passage from FS to CS was confirmed through dedicated observation windows (Fig. 2), water stress was induced. All non-destructive sampling was initiated within 24 hours of the treatment. Subsequently, the plants were allowed to grow for an additional month before being harvested for morphological trait analysis. Preliminary assessments of root exudates, gene expression, substrate-derived volatile organic compounds, and key morphological traits (e.g., shoot/root biomass and length, total leaf area, root system architecture) are shedding light on the molecular and physiological mechanisms underlying both inter- and intraspecific signaling under water stress conditions.



Fig. 1. Experimental setup at day 2



Fig. 2. Roots of *T. patula* L. passed in CS sector

3.4 = Citizen Salad: A Citizen Science Approach to Investigating Plant Leaf Microbiota

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Lettuce is one of the most widely consumed raw vegetables, making it crucial to understand and predict its leaf-associated microbial communities for both plant health and human well-being. While environmental factors play a significant role in shaping plant microbiomes, the impact of plant-specific traits and the origins of microbial diversity remain largely unexplored.

To address these knowledge gaps, we launched a citizen science experiment, partially funded through crowdfunding. The campaign attracted 144 donors and engaged 100 participants across Italy in growing two lettuce varieties and collecting soil, environmental, and leaf samples. The project involves a diverse range of contributors, including schools, eco-museums, urban gardens, and botanical gardens.

Experimental design: in April 2025: each experimenter received 2 cv of lettuce (*cv Parella*, *cv Regina di maggio*) in seed to grow according to their experience and space availability. A field sheet reports all the cultivation specifications. Approximately 60 days after transplanting, samples of leaves, soil and environment were collected. Bacterial communities will be analysed by 16S rRNA amplicon sequencing to distinguish the contribution of plants, soil and environment to the total microbial community.

By combining public engagement with rigorous, hypothesis-driven research, Citizen Salad seeks to advance our understanding of plant microbiomes while fostering scientific literacy and community participation.

3.5 = Preliminary data on the resupination of three Euro-Mediterranean orchid species

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The flowers of Mediterranean orchids are characterized by a zygomorphic morphology with a horizontally oriented floral axis. This structural arrangement imposes a unidirectional trajectory on pollinators, resulting in greater accuracy and precision of pollination through alignment of reproductive organs and deposition of pollen onto a specific region of the pollinator's body (1). By directing the pollinator along a defined flight path, this floral orientation maximizes pollen-placement precision and overall pollination efficiency (2).

In most Orchidaceae species, horizontal orientation is achieved through resupination, an approximately 180° rotation of the floral axis caused by twisting of the peduncle, rachis, pedicel, ovary, or even the corolla. This rotation repositions the labellum (the modified median petal) ventrally, enabling it to function as a landing platform for pollinating insects or, in some taxa, as a structure that enhances the attraction and temporary retention of floral visitors.

Although resupination has been documented in nearly all wild European orchids, few studies have thoroughly investigated the underlying mechanisms, specific modes of action, and timing across different species.

In this study, we examined resupination in three Euro-Mediterranean orchid species, *Anacamptis morio* (L.) R.M.Bateman, Pridgeon & M.W.Chase; *Anacamptis papilionacea* (L.) R.M.Bateman, Pridgeon & M.W.Chase and *Ophrys lutea* Cav., using morphometric and chronometric analyses under laboratory conditions. Through direct observations and sequential photographic documentation, we measured the angle of floral rotation, characterized the dynamics and duration of the process, and identified the onset and completion of flower torsion.

These findings enhance our understanding of the mechanisms and temporal patterns of resupination in Euro-Mediterranean orchids and provide a basis for developing more effective conservation strategies.

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3.5 = Differences in nectar chemistry and pollinator communities between populations of non-obligate serpentinophytic *Onosma* spp. (Boraginaceae: Lithospermeae)

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Serpentine soils are chemically stressful environments for plant growth and reproduction, since they contain low nutrient concentrations and high levels of heavy metals such as nickel, chromium and cobalt, which are known to be potentially phytotoxic. Despite these harsh conditions, serpentine soils can support specialized plant species which are tolerant to (or even hyperaccumulate) heavy metals; these species may be restricted to serpentine substrates or occur also in other edaphic conditions. It is known that these specialized species can translocate heavy metals from soil to their vegetative tissue, and recent studies under controlled greenhouse conditions have shown that metals can be transported also to floral nectar (1). Since floral nectar and pollen are the main food sources for pollinators, and because exposure to metal concentrations can affect cognitive performance and foraging behaviour, and even lead to elevated mortality in honeybees, bumblebees and some wild bee species (2), a deeper understanding of the relationships between heavy metal translocation, pollinator visitation and plant fitness is required. The aim of this study is therefore to understand if heavy metal translocation from serpentine soil to nectar occurs under natural conditions, to highlight if the stressful conditions of serpentine soil could affect nectar quantity and quality in terms of sugar, amino acid and biogenic ammine concentrations, and to analyse potential consequences in the pollinator community. Plants of the genus *Onosma* (Boraginaceae: Lithospermeae) are suitable for this study since some of its species are serpentine tolerant, and populations growing on serpentine and non-serpentine substrates can be found in quite proximity. They also produce a considerable amount of nectar and are pollinated both by generalists and specialized wild bees. Comparing populations of plants growing on serpentine and non-serpentine soils in terms of nectar chemistry and pollinator communities could shed some light on this matter.

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3.5 = From flowers to molecules: secondary compounds in plant-pollinator interactions

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About 60% of all angiosperms produce floral nectar to attract their pollinators. It is a floral secretion with a complex chemistry. Although sugars and amino acids are the most abundant solutes, in recent years other substances present in lower concentrations have been highlighted (2). Among these, several types of secondary compounds have been recognized to have important roles in shaping plant-pollinator relationships. Our contribution focused on two types of secondary compounds, namely non-protein amino acids and biogenic amines. We demonstrated that γ -aminobutyric acid (GABA) can prolong insect survival, and β -alanine to stimulate fly activity at nectar-like concentrations in *Osmia bicornis* and *Bombus terrestris*, respectively. Concerning biogenic amines, it was demonstrated that nectar-like concentrations of tyramine decrease the time spent by insects visiting flowers. Our findings, combined with those obtained by other researchers (3), point to the role of nectar secondary compounds in potentiating the pollination activity of insects. We also demonstrated that in this scenario, nectar-dwelling microbes may also have a role in affecting the concentration of secondary compounds and thus insect foraging behaviour revealing a tripartite interaction.

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3.6 = Assessing restoration success through plant–pollinator networks in lowland wetlands

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The low-alkaline fens of Friuli Venezia Giulia represent a precious complex of lowland ecosystems that suffered from heavy alterations due to human activities throughout the XX century. In these areas, most of the natural fens, grasslands and spring ponds got replaced by intensive crop farmland. This caused an extreme simplification of the landscape and a loss of many endemic and characteristic animal and plant species. From the early 2000s up to 2014, the LIFE program sustained the effective restoration of more than 40 ha of natural habitats, recreating the waterlogging gradient, increasing landscape complexity and reintroducing lost species. This work aims to evaluate the success of the ecological restoration actions on the plant-pollinator networks. During the growing season of 2024 (May to August), we sampled wild pollinators visiting flowers in four wetlands in Friuli Venezia Giulia. We considered the restoration of two habitat types, low-alkaline fen and grassland, comparing the 9 restored and natural patches, for a total of 36 sampling sites (9 x 2 x 2). There we conducted net sweeping in 200 m² transects for a total of 30' each, two times throughout the sampling season. We collected wild pollinators mainly belonging to *Apoidea* superfamily and *Syrphidae* family, recording the plant species every individual visited, and the overall plant diversity in plot areas centered on each transect. Restored fens, grasslands and natural grasslands all had similar pollinator richness, while the natural fens showed significant lower values. Nevertheless, when considering intra-group β -diversity, natural fens showed the highest value, followed by restored fens, natural grassland and restored grassland. These findings suggest that old growth natural fens host few but highly adapted pollinators, mainly syrphids belonging to genera *Melanostoma* spp., *Eristalis* spp. and *Helophilus* spp. Pollinator communities visiting the other habitats, i.e. natural and restored grasslands and restored alkaline fens, are shown to be richer but less distinct, possibly reflecting a still ongoing process of differentiation of the vegetation¹; in facts, the most visited flower species were *Potentilla erecta* (L.) Raeusch., *Centaurea nigrescens* Willd. and *Ranunculus acris* L., three species with broad ecological niches that are commonly found in early successional stages; moreover, pollinators in low-alkaline wetlands visited very few habitat-specific plant species (eg. *Gladiolus palustris* Gaudin, *Blackstonia perfoliata* (L.) Huds, *Mentha aquatica* L.), despite their presence. This can possibly signify that specialization and interaction complexity in pollination networks require longer periods of time to be restored. Nonetheless, the analysis of pollination network, provided insightful knowledge for a conservation perspective: when analyzing the nestedness² index, restored grasslands show the highest value, suggesting a more complex web of interactions³. Lastly, in the restored habitat subset, the index decreases with age only in grassland habitats, implying a potential progressive specialization of the flora, with a progressive loss of redundancy of the interactions.

These findings help to build a more complete picture of the present restoration status of the area, helping to disentangle the role of time in re-establishing inter-*taxa* relationships, and allows to depict a possible evolutive trajectory of the studied communities.

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3.6 = New Bees, Old Tricks: When *Ophrys* orchids and naïve pollinators meet in novel habitats

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Large populations of rewarding plants tend to attract more pollinators, leading to higher visitation rates and enhanced pollination success. Conversely, small populations may experience insufficient pollen transfer, resulting in reduced seed production. Plant density plays a crucial role in influencing pollinator behavior; pollinators are more likely to move between closely spaced individuals in dense populations due to shorter flight distances. In contrast, in lower-density populations where plants are more spread out, pollinators may find it more challenging to transfer pollen effectively. Nonetheless, these general patterns can be complicated by deceptive flowers. Orchids, for example, often feature a significant number of unrewarding species. The pollination strategies employed by unrewarding orchids include both food deception and sexual deception, utilizing olfactory and visual cues. Food deception involves mimicking the floral signals typically associated with rewards and is pollinated by female bees, while sexual deception attracts male pollinators by imitating the pheromones or appearance of their female partners.

For food-deceptive orchids, it was predicted and found that pollination rates would be higher in low-density environments with less competition from rewarding counterparts. In contrast, male pollinators of sexually deceptive orchids are not expected to behave like female bees. We predict that clustered groups of sexually deceptive orchids will be more appealing than isolated or sparsely clustered individuals, as they can emit a more intense sexual fragrance that draws in male pollinators. Furthermore, we hypothesize that fruit set will be greater in areas where orchids have not previously been established compared to areas where they already exist. To test this hypothesis, we established both high- and low-density artificial populations of *O. sphegodes* in locations devoid of orchids and subsequently compared the fruit set of these artificial populations to that of natural populations of the same orchid. The analysis shows no differences in the fruit ratio between low and high-density groups of *Ophrys* plants, both in artificial and natural populations. Nevertheless, we found a significantly higher pollination success in the artificial populations (where the orchids did not occur before) compared to those in the natural population, regardless of plant density. We speculate the introduction of sexually deceptive orchids into a novel environment enhances the frequency of interactions between pollinators and plants as the local male bees are naïve concerning the deceptive flowers. In contrast, in natural populations, male bees quickly become accustomed to the presence of the orchids and avoid them. Accordingly, in the natural populations, we found that the early-blooming orchids (i.e., those that first encounter the naïve male bees) have higher pollination success than late-blooming ones.

4.3 = Towards a national aggregator of botanical data in Italy

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The fragmentation of botanical data across multiple datasets and repositories represents a significant barrier to biodiversity research and conservation planning. While individual datasets provide valuable insights, merging data from different sources (a challenging and time-consuming task) is often required to perform complex analyses.

Data aggregators, by linking different kinds of information together and providing a single access point to the user, are thus pivotal in overcoming such limitations.

In Italy, the web portal FlorItaly hosts checklist data for vascular plants, covering scientific names, synonyms, and distribution details. Building on this infrastructure, we are expanding the current system and creating a true botanical data aggregator for the country.

To achieve such goal, we are working to integrate:

- occurrence data from herbarium specimens, which have been digitized through the framework of Spoke 7 of the National Biodiversity Future Center (NBFC),
- DNA barcoding sequences and phylogenetic information linked to voucher specimens,
- secondary metabolites data from scientific literature and from investigations carried out within the framework of the NBFC,
- morphological and functional trait data from the resources of the *Dryades* project.

All these data will be made available in the web portal through query interfaces. Furthermore, they will also be downloadable in standard formats and accessible through public APIs in programming environments such as R and Python.

4.5 = Reflections (*Inventio/Commentatio*) on Common Western Naming Practices for the Horticultural Groups of *Rosa* L.

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This paper examines the equivalence of horticultural names commonly used in Western countries for groups of *Rosa* L., tracing their historical development. It highlights how designations emerged through the intersection of botanical and horticultural practices, and how these names reflect both taxonomic intent and cultural tradition. While botanical nomenclature follows internationally recognized codes, horticultural naming - especially before the 20th century - often evolved independently, driven by marketing, affective associations, or homage. Roses have been extensively manipulated, and the classification of cultivars is complicated by uncertain ancestry and overlapping morphological traits. Historically, horticultural classifications attempted to group roses based on key characteristics and presumed parentage, though this approach had its limitations. The establishment of classes such as “Miscellaneous OGRs”, where OGRs stands for Old Garden Roses, reflects the persistent challenge of fitting all cultivars into rigid taxonomic schemes. From Redouté and Thory’s *Les Roses* (1) to the commercial classifications of 19th-century nurserymen, efforts to organize roses for identification and trade grew alongside the horticultural market. Early works by Vibert (2), Buist, and Pucci laid the foundation for group-based classifications still in use today. In the 20th century, both professional breeders and international organizations, such as the World Federation of Rose Societies, proposed revised classification systems, including the 1979 tripartite division into wild, old garden, and modern roses. However, no universally accepted horticultural classification exists to date. National traditions, linguistic ambiguities (such as with the terms “Thea” and “Climbing”), and the blending of scientific and commercial goals continue to complicate standardization. The American Rose Society’s (3) evolving framework, though widely adopted, coexists with parallel systems reflecting regional and sectorial priorities. Ultimately, the naming and classification of roses remain dynamic fields that mirror the intricate relationship between science, commerce, and culture. The diversity of systems illustrates both the richness of the rose’s legacy and the ongoing effort to balance historical depth, botanical accuracy, and practical use.



Fig. 1. *Rosa* × *thea* Savi (= *Rosa* × *odorata* (Andrews) Sweet var. *odorata*.
From Savi (1822)

1) Redouté P.J., Thory C.A., 1817. *Les Roses*. F. Didot, Paris. 283pp

2) Vibert J.P., 1831. *Observations sur la nomenclature et le classement des roses, suivies du catalogue de celles cultivées*. M.me Huzard, Paris

3) American Rose Society, 2018. *Handbook for selecting roses*. ARS, Shreveport (USA)

4.5 = Slovenian botanical treasures – Slovenian oldest herbarium books

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The Slovenian Museum of Natural History is the one of the successors of the Provincial Museum of Carniola, founded in 1821, which collects, preserves, studies and presents the natural heritage of Slovenia. Among its collections are also botanical ones, especially old herbaria. The first herbaria in the world, in the modern idea of collecting true plants, were formed in the 16th century in Italy. They have been made as herbarium books and often called “*hortus siccus*” (dry/dried garden) or “*herbarium vivum*” (1). The oldest known herbarium from Slovenia is an herbarium book made by a physician from Ljutomer (NE Slovenia), Janez Krstnik Flysser in 1696 (2). However, the museum also keeps an herbarium book from the legacy of Jožef Kalasanc Erberg, that hasn’t been analyzed and properly presented before. The Flysser Herbarium is a book with leather-bound wooden covers and contains 993 relatively well-preserved plants labelled with Latin names and in some cases added German ones (Fig. 1). It doesn’t specify the location at which plants were gathered, though botanists presume most were collected on the territory of the former Venetian Republic, in Italy and areas under Italian influence, while some are ornamental and cultivated species. The book later belonged to Karel Zois (1756–1799), who was also a botanist and wrote the herbarium list of plant species. As a part of the Zois bequest, the herbarium was transferred to the library of the Ljubljana Lyceum, which in turn gave it to the Provincial Museum of Carniola in 1838. With the bequest of baron Erberg another herbarium book became in the possession of the mentioned museum. Josef Kalasanz von Erberg (1771–1843) was a senior official on the Austrian court, also a historian and a patron, who owned an artwork collection, a rich library and several natural objects. Following the example of his friend Sigismondo Zois (1747–1819), who most likely provided Flysser’s herbarium to his brother Karl, he also owned a Renaissance herbarium book. On the first page, an *ex libris* indicates that the owner of the herbarium book was J. K. Erberg and the name Caroli Weichhart, student of medicine, and most likely the author of the herbarium, is crossed out. On 100 pages, 389 specimens of plants are attached, decorated with yellow and red vases (Fig. 2). The use of various vases, as decorative elements in herbaria, was popular especially at the end of the 17th and in the first half of the 18th century and is probably related to the fact that plant collections were perceived as dried gardens, where plants (albeit on paper) were placed in vases (e.g. Georg Clifford’s herbarium and Zierikzee herbarium; 4). Most of the plants in the herbarium grow on the territory of Slovenia, with only a few cultivated and ornamental species. However, there are also plants that are rare in Slovenia or don’t grow there (e. g. *Primula glutinosa*). Therefore, most likely plants for the herbarium were collected in the area of the Habsburg Monarchy, for example in present-day Austria and Slovenia. They are labelled with pre-Linnean plant names, which also places the origin of the herbarium in the times before Carl Linnaeus introduced the use of binomial nomenclature in science – in the first half of the 18th century.



Fig. 1. Flysser’s Herbarium. Photo: David Kunc

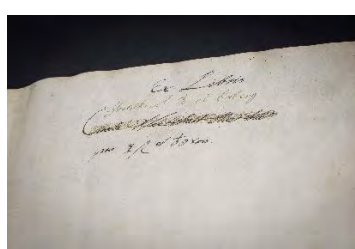


Fig. 2. Erberg’s Herbarium with vases. Photo: David Kunc

1) T. Van Andel (2017) Open the treasure room and decolonize the museum. Inaugural lecture delivered on the occasion of the acceptance of the position of Special professor of the Clusius chair of History of Botany and Gardens at Leiden University. Page 4

2) N. Praprotnik (2015) Botaniki, njihovo delo in herbarijske zbirke praprotnic in semenk v Prirodoslovnem muzeju Slovenije / The botanists, their work and herbarium collections of vascular plants in the Slovenian Museum of Natural History. Scopolia 83/84: 414 pp

3) A. Offerhaus, E. De Haas, H. Porck, A. Kardinaal, R. Ek, O. Pokorni, T. van Andel, (2021) The Zierikzee Herbarium: contents and origins of an enigmatic 18th century herbarium. Blumea, 66 (1): 1–52

4.5 = The Biella Paradox: The Resilience of Plant Foraging in a Post-Industrial Pre-Alpine Area of NW Italy

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This study investigates the continuity and transformation of wild plant foraging practices in the Biella province of Piedmont, northwestern Italy, over the past fifty-five years. By conducting ethnobotanical fieldwork with 15 local informants using semi-structured interviews and snowball sampling, the research documents the current use of 82 wild plant species for food and herbal teas. These findings are compared to a seminal ethnobotanical survey from 1970 (published in 1992), which recorded 93 wild food species in the same area. The comparison reveals both continuity and change, with a decrease in the overall number of species used, but notable consistency in the most represented botanical families, Asteraceae, Polygonaceae, and Rosaceae, across both periods. The enduring presence of these families highlights the persistence of key traditional foraging knowledge. The relative resilience of plant use traditions in Biella contrasts with steeper losses observed in other European regions and may be attributed to several interrelated factors. These include the geographic marginality of the region, which may have shielded it from rapid socio-cultural changes, the economic and demographic consequences of the textile industry collapse, which reinforced subsistence practices, and strong local traditions such as home gardening and communal land use. The study illustrates how environmental, cultural, and economic conditions interact to support the survival of traditional ecological knowledge. It underscores the importance of recognizing and integrating such knowledge into regional sustainability efforts, food security strategies, and cultural heritage preservation initiatives, especially in the context of ongoing environmental and social transformation in rural and peri-urban areas.

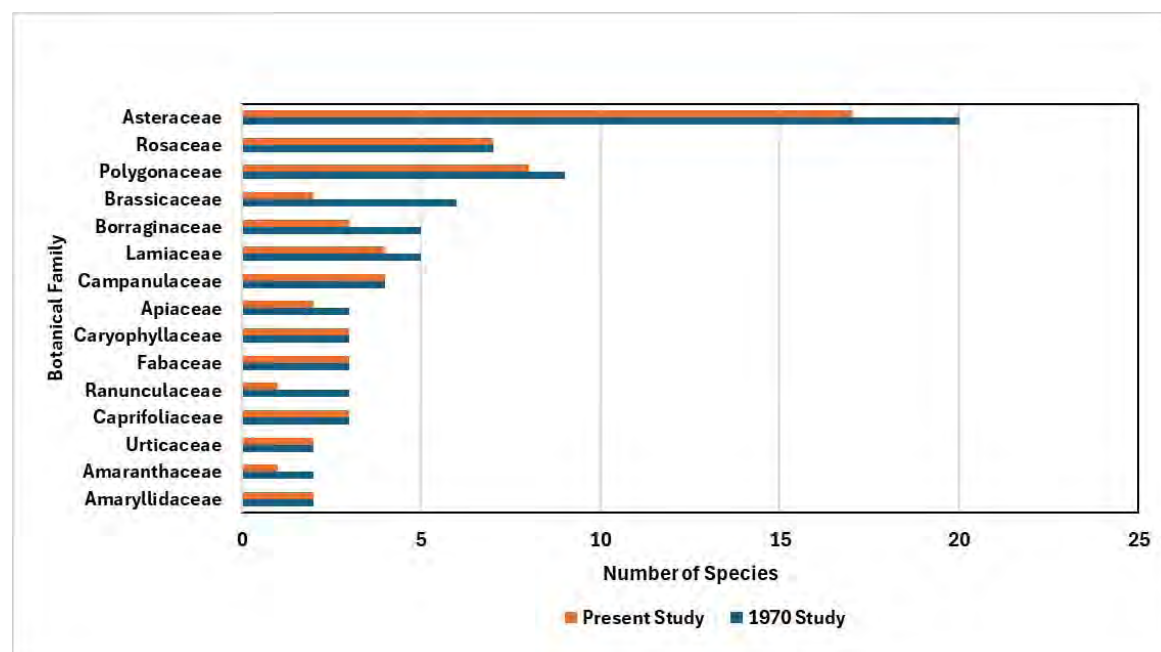


Fig. 1. Comparison of the most represented botanical families in the studies conducted in 1970 and 2025

1) Sella, A. 1992. Flora popolare biellese: Nomi dialettali, tradizioni e usi locali. Edizioni dell'Orso, Alessandria

4.6 = An update about the massive data production by scanning of the main italian herbaria as a PNRR project of the CN5

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In the context of the project CN5 (PNRR) and National Biodiversity Future Center (NBFC), one of the most important activity of spoke 7 is the massive scanning at high resolution of a large part of the herbarium specimens conserved in Italy. The center of this activity is the Herbarium Centrale Italicum (HCI) in Florence, being this institution the largest repository of herbarium specimens in Italy, with millions of exsiccata. To reach this goal, the activity established a partnership with the Dutch company Picturae, owner of equipment specifically designed to this goal, with an average weekly production of 60000 images.

After the first phase in Florence, centered on the HCI, other italian herbaria are being invited to participate to the effort, by sending their specimens to Florence for digitization arriving at 4,250,000 specimens, most of them of the Mediterranean region, and associated metadata with the aim to make millions of specimens available for investigation at a distance. The digitization of a large number of herbarium specimen will produce a large database of high resolution images and data that are and will be always a useful source of information for the investigation of biodiversity (1). The images (jpg format) are being transferred to a CINECA repository for testing methods of artificial intelligence for investigation of high numbers of specimens, using both the plant material and the metadata obtained from the label (Fig. 1).

Ente finanziatore: Progetto finanziato dall'Unione Europea - NextGenerationEU – Piano Nazionale Resilienza e Resilienza (PNRR) - Missione 4 Componente 2 Investimento 1.4 – Avviso N. 3138 del 16 dicembre 2021 rettificato con D.D. n.3175 del 18 dicembre 2021 del Ministero dell'Università e della Ricerca



Fig. 1. A fern specimen



Fig. 2. Multiple specimens herbarium specimen. Bar = 2 mm

1) Heberling, J. M., Prather, L. A., & Tonsor, S. J. (2019). The changing uses of herbarium data in an era of global change: an overview using automated content analysis. *BioScience*, 69(10), 812-822

4.7 = Multitaxon floristics and botanic collections in the Natura 2000 site IT 3230042 Torbiera di Lipoi (Feltre – Belluno)

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As part of the “Bando Habitat 2020 – Progetto di rigenerazione e valorizzazione della Torbiera di Lipoi” funded by the Cariverona foundation, an investigation was started to characterize the vascular flora and the bryo-lichen biota of the Natura 2000 site IT3230044 called “Torbiera di Lipoi” located in the Municipality of Feltre (Province of Belluno; surface area: 65 ha; altitude: 320 m). It is a low alkaline fen, known since the 1970s for its floristic importance, surrounded by a semi-natural agro-forestry landscape.

As regards the vascular flora, the research was developed along two main lines: a) aggregation of existing floristic data as a basis for comparison; b) a new floristic investigation aimed at verifying any changes over a period of fifty years. The bryo-lichenological investigation instead focused on the oak-hornbeam woods bordering the fen where there are tree individuals (especially *Quercus robur* L. and *Carpinus betulus* L.) of considerable size and a lot of dead wood. These woods are crossed by small temporary streams, with a stony bed, which flow towards the fen.

During the field work, specimens were collected for the preparation of voucher specimens both for vascular plants and for bryophytes and lichens. This material formed the basis for the creation of a digitalized collection that is available on the project website (<https://comune.feltre.bl.it/torbiera-di-lipoi/>).

Overall, 319 taxa of vascular plants were recorded. This checklist provided the basis for the creation of an interactive identification guide available from Dryades (<https://dryades.units.it/home/index.php>) and from the project website.

Results indicate that, despite natural reforestation triggered by the abandonment of the traditional management, most of the fen-related vascular species still occur, including red-listed species. Moreover, alien species are still rare, occurring in small scattered populations. However, the strong reduction of the fen surface during the last twenty years suggests that the current vascular flora may reflect a previous situation in which fen-related habitats were much larger than today.

Despite the fact that the bryo-lichen biota is still likely non exhaustively explored, we inventoried 27 bryophyte taxa (6 liverworts and 21 mosses) and 22 lichen taxa. Most of them are linked to the two prevalent tree species (*Q. robur* and *C. betulus*) and partly to dead wood. The lichen biota is mainly composed of species with a mild-temperate to temperate-montane distribution, attributable to the Graphidion scriptae communities. Half of the species are characterized by trentepohlioid photobionts, such as *Pyrenula nitidella* (Schaer.) Müll. Arg. and *Opegrapha vermicellifera* (Kunze) J.R. Laundon. In the stony bed of the streams luxuriant populations of the bryophyte *Fontinalis antipyretica* Hedw. Were found, as well as the lichen *Verrucaria praetermissa* (Trevis.) Anzi, which is currently known in Veneto only from this location.

Overall, the study provided the scientific basis for drafting guidelines for conservation and monitoring interventions that will be implemented within the project with the aim of maintaining in the long-term the remaining fen areas.

4.8 = Integrated taxonomy of herbarium specimens support recognition of new species of *Austroepiphloea* (Halymeniales, Rhodophita) from Madagascar

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The Atimo Vatae expedition, held in the spring 2010, was carried out by the combined effort of the Muséum National d'Histoire Naturelle (MNHN, Paris), the Institut d'Haliéutique et des Sciences Marines, University of Toliara (IH.SM), and the Wildlife Conservation Society (WCS) Madagascar Programme (1). Through comprehensive sampling, it explored the coastal marine fauna and flora from Androka to Fort Dauphin, in southern Madagascar, a cold water region where the algal flora is luxuriant and where endemism is supposedly high. All the algal samples are housed in the cryptogamic collection (PC) in the Herbarium of Muséum National d'Histoire Naturelle (MNHN, Paris) as silica-dried and herbarium samples and were subjected to a preliminary DNA barcoding screening.

In collaboration with the Muséum National d'Histoire Naturelle (MNHN, Paris), our research group was entrusted with the study of Halymeniacean samples. The first result was the description of two new species of *Yonagunia* S. Kawaguchi & M. Masuda (2, 3). In the present study, we report on seven herbarium samples identified as two related taxa, which were analysed by DNA barcoding, phylogenetic and morpho-anatomical analysis. The collected data support the description of two new species (Figs. 1 and 2) of the genus *Austroepiphloea* Molinari, Sánchez Ocharan & Guiry (provisionally named *A. sp1MAD* and *A. sp2MAD*), a genus considered monospecific and endemic to Australia (3). Our in-depth analysis of the Museum samples consists of two principal study; the production of nuclear, mitochondrial and plastid gene sequences (*coxI*, *rbcL*, *LSU*, *psbA*) for phylogenetic analysis with a combined multigene approach, and morpho-anatomical study of herbarium specimens, which allowed us to observe both the vegetative and reproductive structures, from a gametophytic cystocarpic specimen and a sporophytic specimen of *A. sp1MAD*.

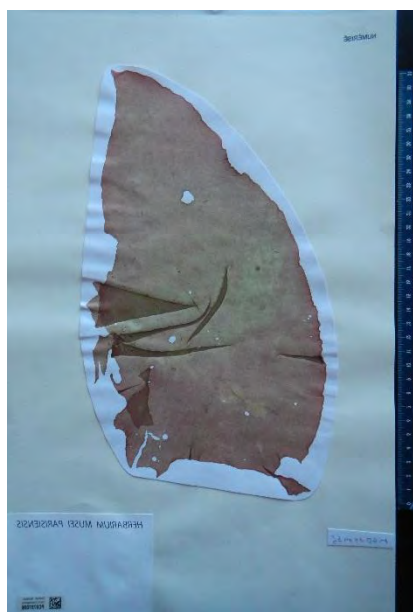


Fig 1. *A. sp1MAD* (PC0737006)



Fig. 2 *A. sp2MAD* (PC0166101)

1) P. Bouchet, P. Perez Bouchet, T. Perez, L. Le Gall (2010) <https://doi.org/10.17600/10110040>

2) A. Manghisi, M. Morabito, G.H. Boo, S.M. Boo, C. Bonillo, O. de Clerck, L. Le Gall (2015) *Cryptogam., Algol.*, 36 (2), 199-217. <https://doi.org/10.7872/crya.v36.iss2.2015.199>

3) E.A. Molinari Novoa in M.D. Guiry & G.M. Guiry (2023) *AlgaeBase*. <https://www.algaebase.org> (searched on 05/05/2025)

4.9 = From integration to innovation: how unified botanical collection management redefined museum policy and practice

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This work presents the reorganization and unified management of the living botanical collections at MUSE - Science Museum of Trento. Previously dispersed across several geographically and administratively distinct units—including an Alpine Botanical Garden, tropical greenhouses focused on African flora, a public ornamental garden, and a recently established wetland biotope—the collections were brought under a single management framework since 2023.

A key step in this integration was the creation of a unified accession dataset. This required the homogenization of plant nomenclature and accessory data, which had previously reflected the diverse expertise and historical contexts of the different sub-units. The development of this dataset highlighted significant heterogeneity in the collections' original aims and structures. In response, cross-cutting guidelines were formulated, clear functional roles for each sub-collection were defined, and a new institutional position statement was issued, aligning the collections with the museum's broader strategic vision.

Concurrently, a comprehensive restoration of the collections was implemented. Key actions included updating and standardizing missing data, ensuring compliance with national and international regulations, donating incongruent accessions, and establishing a new educational collection.

Integration with BGCI platforms (PlantSearch and IndexSeminum) improved nomenclature standards and revealed residual data inaccuracies. Staff training and dataset merging expanded the number of species available through the Index Seminum exchange network (previously limited to the Alpine Botanical Garden) boosting international visibility and material requests. This integration also enabled new research avenues, including preliminary studies using BGCI climatology tools to explore the role of living collections in climate monitoring. Additionally, the redefinition of collection objectives has supported participatory projects in social inclusion, education, and art-science collaboration.

Managing this increasingly complex dataset necessitate the acquisition of specialized collection management software, expected to further enhance research potential and data-driven exploration.

Collectively, these initiatives have not only optimized collection management but also redefined the functional paradigm of the botanical collections in alignment with modern scientific and cultural development trajectories.

4.9 = A truffle cultivation museum for scientific culture and cultural inheritance

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Truffle hunting and extraction, and the body of knowledge concerning them, are often held by the older generations living in small villages in rural areas. For the populations of inland areas, truffles are not only a symbol of Italy in the world, but their importance is also practical: economically, truffles provide income for people living in hilly and mountainous areas, among the most disadvantaged (1). Following the decline in natural truffle production, truffle cultivation quickly became an investment opportunity for farms in a multi-functional approach (2). Given the economic, agricultural, and cultural value of this fungus, it is essential to safeguard the traditional knowledge associated with it and integrate it with new scientific discoveries. Following the success of the project “Culture and territory: exhibitions and conferences on the biology, ecology and cultivation of truffles as a means of spreading culture in the Umbrian territory, PANN12_00267,” the idea of a permanent scientific exhibition in Fratta Todina (PG) was born. Its realization was made possible through the submission of a project led by the Municipality of Fratta Todina, which received funding from the GAL Media Valle del Tevere, under measure 19 - sub-measure 19.2 “Redevelopment of public landscape and historical-cultural heritage in the rural areas and villages of the Media Valle del Tevere,” part of the 2014 – 2020 Rural Development Program of the Umbria Region. The goal is to raise awareness among younger generations and the entire community, to foster sustainable cultural development where citizens play an active role. For this reason, the museum was born from the dialogue between sector experts, freelancers, and associations such as the Pro Loco of Fratta Todina and the University of Perugia (2). Scientific museums not only can foster awareness and responsibility toward nature through environmental conservation education but also provide social benefits. Indeed, cultural heritage can act as a social bond that contributes to human development and cultural diversity appreciation (3). The “Tuber Tiber” museum was conceived by Dr. Paola Ballario and developed by Leonardo Baciarelli Falini. The structure is housed in the historic Palazzo Rivelloni in Fratta Todina. The exhibition features illustrative panels and touch screens presenting truffle potential mapping, vegetation and geological charts, as well as truffle ecology and biology, species morphology, and peridium, gleba, and spores structure. Visitors can also explore ectomycorrhizae with the magnification of mycoclena and peritrophic hyphae and learn about truffle hunting. Interactive learning is also available through cases containing various soil types, sections of symbiotic tree trunks, fruits and seeds of major truffle-hosting plant species, and truffle plants. Visitors are also provided with a stereo and optical microscope. The exhibition will be complemented by excursions, seminars, specialized courses on truffle cultivation and much more (2). The museum is designed for everyone, but especially for young people and students, so that they can understand what scientific research is and what its practical implications are. The hope is that visitors will become aware of the truffle biodiversity in their territory and of the cultural heritage they can represent.



Fig. 1. A lesson at the museum



Fig. 2. Illustrative panels and touch screens

1) Bragato G., D. Mosetti, P. Turpaud, G. M. N. Benucci, D. Donnini (2022). *Applied Soil Ecology*, 178, 104565

2) Falchetti E., D. Grohmann, A. Barili, M. Maovaz, S. Gentili (a cura di) (2022). *Atti XXX Congresso ANMS*, Perugia 5-8 ottobre 2021, *Museologia Scientifica Memorie*, dicembre 2022 n. 22

3) Falchetti E., B. Utzeri (a cura di), (2013). *Associazione Nazionale Musei Scientifici*. ISBN 978-88-908819-0-9

4.9 = Enhancing the collections of the Botanical Museum at the University of Padova: ongoing projects and future perspectives

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Natural history collections provide a huge amount of information, representing a key element in the study of biodiversity and its changes as well as in the reconstruction of the history of a collection and its collectors. Among these, botanical collections, including herbaria, have been increasingly studied in the last few years thanks also to technological advancements. In particular, museums and botanical gardens around the globe (e.g. Royal Botanical Gardens–Kew) have started digitizing their herbarium collections. Digitization represents the first step to make the collections and their data more accessible and exploitable both for researchers and non-experts. The Botanical Museum of the University of Padova, located in the Botanical Garden, hosts the Herbarium Patavinum (PAD) and other botanical collections. The Herbarium, established in 1835 by director Giuseppe Antonio Bonato (1753-1836), houses today around 600.000 specimens. The botanical collections in Padova are involved in several projects, in order to enhance their potential both in terms of historical and scientific resources. First of all, the Herbarium specimens are currently being digitized as part of a large-scale digitization process promoted in Italy by the National Biodiversity Future Centre (NBFC) coordinated by the University of Padova and going on at the Botanical Museum of the University of Florence. The digitization will provide a more precise number of the specimens as well as more information about the taxa preserved and the history of the collections (e.g. name of the collectors, time and place of the collection). Another digitization project is taking place in situ, involving the Algarium of Achille Forti (1878-1937). Specifically, the digital image acquisition is currently happening and it will be followed by the data extraction from the labels in order to put together a complete database. The work done so far has significantly increased the estimated number of specimens, placing the PAD algae collections among the largest in Italy. In addition, some botanical collections, specifically the Algarium of Forti, the cecidological collection of Alessandro Trotter (1874-1967) and the herbarium collection of Silvia Zenari (1895-1956), are under investigation.

The ongoing research on Forti's Algarium and his scientific activity, carried out by integrating the collection's data, the archival documents, and the scientific literature, aims to reconstruct the history of the collection and to place Forti within the studies and the community of algologists between the 19th and 20th centuries.

The study of Trotter's collection provides valuable data for analysing the relationship between host plants and gall-forming organisms, as well as the diversity of galls. The aim of the project is twofold: to create a dataset in order to map collection sites and identify sampling biases, and to explore the emergence of 'cecidology'—the science of galls. This work has also led to the creation of an exhibition at the Botanical Museum to present Trotter's studies and his collection to the public.

Finally, the study of Zenari's specimens focuses on the reconstruction of her botanical expeditions in Northeast Italy and abroad as well as on the characterization of her collection, in order to go into detail about its value in terms of biodiversity conservation. The method involves combining data from archival research (e.g. archives of University of Padova) and online open repositories (e.g. GBIF) as well as getting in touch with curators of herbaria (e.g. Italian Central Herbarium) and Zenari's heirs.

All of these projects will enable us to address relevant current issues such as the colonial legacy of botanical collections and its impact on the present as well as the role of women.

In Padova, the convergence of digitization with current and future studies will foster the emergence of new narratives and methods to engage the public as well as to raise awareness about the value of the botanical collections as cultural and natural heritage. In summary, our work aims at i) understanding the history of the botanical collections and their collectors, ii) making the collections and their data accessible, iii) combining history of science and botany (interdisciplinary approach) to study biodiversity, iv) enhancing the botanical collections as historical, cultural and natural resources.

4.10 = Tracing Millennia of Flora and Land Use: Archaeo-Palynological Insights from Southern Italy

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The archaeo-palynological investigations conducted across archaeological sites in Campania, Basilicata, and Sicily allows the reconstruction of millennial patterns of floristic diversity and land management in these regions (1). Sites were extracted from the BRAIN database, and 14 of them were selected for data elaboration based on the availability of pollen data produced by the Laboratory of Palynology and Palaeobotany of Modena (2). Pollen analysis yielded detailed palaeo-floristic lists that reflect the biodiversity, habitats, and ecosystems surrounding the settlements over time. These records provide evidence that palynology, when conducted at high taxonomic resolution, can operate effectively at the floristic level, enhancing our understanding of past plant communities and their diversity. Based on the arboreal/non arboreal ratio of all pollen identified in the spectra, it emerges that the landscapes in the study areas were already open in ancient times, with significant openness during the Roman period. This condition probably reflects both practical and aesthetic land-use choices, evidenced by the rise in plane tree (*Platanus orientalis* L.) and manna ash (*Fraxinus ornus* L.) pollen curves: these trees are known to have been used for their ornamental, functional, and symbolic value. Pollen of cultivated fruit trees and cereals points to more structured agricultural systems during Roman occupation, suggesting a deep human reshaping of the environment. These transformations in the terrestrial landscape find a parallel in changes observed in aquatic ecosystems. A particularly compelling example is the identification of white water-lily (*Nymphaea alba* L.) pollen in Hellenistic-Roman layers in Sicily. Once part of the island's freshwater ecosystems in the 3rd century BC, this species has not been recorded in the region since the 20th century. Its ancient presence and modern disappearance highlight the sensitivity of aquatic habitats to environmental and anthropogenic changes. Furthermore, region-specific pollen indicators reflect differing cultural and ecological paths: urban-agricultural signatures in Campania, Mediterranean-pastoral landscapes in Basilicata, and agro-pastoral systems in Sicily with a strong Mediterranean biodiversity component. These findings underscore the dynamic interplay between human societies and ecosystems across time, and the impact of cultural practices on biodiversity.

Beyond reconstructing past environments, this research contributes to broader reflections on sustainability and conservation. The palaeo-floristic datasets can serve to identify the traditional vocation of a territory to compare with modern condition, offering critical insights into long-term biodiversity loss, resilience, and environmental changes (3). Pollen records—preserved in both archaeological and natural archives—are unique tools to answer not only questions of landscape archaeology, but also to support current conservation strategies for Mediterranean ecosystems facing ecological pressures.

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4.10 = Integrated analysis of pollen and volatile compounds to trace Calabrian honey floral origin: insights into floristic diversity

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Composition and sensory properties of honey are strongly influenced by the plant species visited by bees, establishing a direct link between honey characteristics and its geographical origin. Characteristics of honey produced in a specific geographical area are primarily determined by local flora and climatic conditions, as flowering periods and nectar availability can vary significantly for the same plant species across different regions.

Chemical and melissopalynological profiles are useful to determine both the botanical and geographical origin of honey: pollen extracted from honey represents a non-chemical analytical method, while volatile compounds -such as terpenes, norisoprenoids, and other aroma-active molecules- provide chemical signatures linked to the floral identity of the honey.

The traditional method for determining the floral and geographical origin of honey is melissopalynology, which focuses on pollen analysis. In addition, the analysis of volatile organic compounds can reflect differences in their profiles, providing valuable indicators for botanical and geographical discrimination (1).

Calabria has been recognized for its high potential in beekeeping, with 47.8% of its territory considered highly suitable for honey production (2), due to its favorable climate and rich floristic biodiversity. However, the composition of Calabrian honeys, in terms of pollen content and volatile profile, has not yet been described. This study aims to characterize honeys from four natural parks of Calabria (a wide region in Southern Italy): Pollino National Park, Sila National Park, Aspromonte National Park, and Serre Regional Park, protected areas with unique pedoclimatic and botanical features. The characterization is based on both melissopalynological and volatile compound analyses, used as botanical fingerprints, and analyzed through

multivariate statistical tools to explore possible correlations among pollen profiles, volatile composition, and production areas. A total of 74 honey samples (produced during the 2023 and 2024 beekeeping seasons) were collected, mainly labeled as wildflower and chestnut honeys. Pollen analysis (3) was performed to identify characteristic pollen associations. The chestnut honeys produced during 2023 were submitted to volatile analysis via Headspace Solid Phase Microextraction followed by Gas Chromatography–Mass Spectrometry (Musci, Cirlini, & Caligiani, 2024).

One hundred different pollen types belonging to 41 botanical families were identified. Regardless of local origin, all samples showed characteristic regional markers: the consistent presence of *Castanea*, *Sulla*, *Echium*, and species from the *Oleaceae* family. Samples from Serre Regional Park and Sila National Park were subjected to statistical analysis in order to explore potential differences in melliferous flora between the two areas.

The volatile profiles of chestnut honeys revealed compounds typically associated with this floral origin, with minor differences related to the production area. The data obtained may help to distinguish Calabrian honeys from those of other geographical areas and provide a foundation for strategies aimed at promoting and protecting the unique botanical and ecological identity of these landscapes.

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4.10 = Natural Traces in forensic investigations - How pollen imprints can solve crime: Review of the open-access literature on forensic palynology

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This research is developed within the Horizon Europe 2020 MSCA Doctoral Network ‘Natural Traces’ project (Grant number: 101120165) dealing with the use of non-human biological traces in forensic science. Forensic palynology is a branch of palynology, the study of pollen grains, that focuses on its implementation in criminal investigations giving microbotanical evidences. Pollen analysis could provide information on the vegetation cover and representation of different plant species in a crime scene, often providing a direct link between objects, people, and places that may pertain to criminal cases. Non-pollen palynomorphs (e.g., fungal spores, algal elements, other microremains of biological origin) that can be observed on the palynological slides are able to give important and additional information about the ecology of the sampling site. The PhD project is aimed at i) studying existing forensic palynological literature, ii) finding and testing methods used in them and iii) modifying some of these protocols to develop the most effective, simple and not expensive in performance methods of sampling and pollen and non-pollen palynomorphs extraction that will work with the most common types of materials and allow crime scene investigators to implement it into an everyday practice. Within this research project, a literature review has been undertaken; this report presents some information got from the search on the 159 open-accessed literature sources (as of May 2025). The review was made for the articles which were published in the time period from 1971 to 2025. The literature research was made in databases of Scopus, ResearchGate and BASE (Bielefeld Academic Search System) with criteria: "forensic palynology"; "forensic AND palynology"; "forensic AND pollen"; "crime AND pollen"; "crime AND palynology" resulted in 160, 101 and 297 findings respectively. The most actively published years for forensic palynology started in 2006. There were found 58 papers that were based on the real cases, 68 sources that were based on the experiments and 33 articles that were reviews. Real cases were associated mainly with homicide, robbery, drug abuse, contrabands, archaeoforensics, rape, etc. The most published papers were found from the UK, USA, Italy, New Zealand and India. Sampling methods were mainly represented by collecting materials, washing, brushing, swabbing the examined materials and surfaces. The most used methods of chemical treatment of samples were Faegri and colleagues' method (1), Erdtman's acetolysis (2) and von Post's method (3). Further steps of the project will be focused on the methodological aspects of palynological researches studied in the represented literature and their effectiveness of pollen extraction from the different materials.

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3) L. von Post (1916) Einige südschwedischen Quellmoore. Bull. Geol. Inst. Univ. Upsala, 15, 219-278

4.10 = Holocene climate-vegetation dynamics within the Mediterranean biodiversity hotspot - insights from an EU priority habitat: the Lago Lao case study (SW Alps)

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Understanding long-term ecological responses to climate variability is critical for informing conservation strategies, particularly in regions of high biodiversity and climatic sensitivity such as the Mediterranean mountains. In this context, we present the Lago Lao case study - a sphagnum peat bog (7110* EU priority habitat) in the Ligurian Alps Regional Natural Park (NW Italy). The site is in a biodiversity-rich area, climate-sensitive, and with a human history dating back to the Pleistocene (1). Pollen data are compared with other available records in the area, integrated with independent paleoclimatic curves at regional scale, simulated local climatic variables, and documented global events/periods, to obtain insights into the ecological dynamics in relation to climate and human influence over the last ~11,000 years. The results highlight the role of increasing warming, during the Younger Dryas-Early Holocene transition, in driving the succession from cold-adapted steppe vegetation (*Artemisia*, Poaceae wild grass group, *Aster* type) to the colonization of heliophilous, cold-tolerant pioneer trees (*Pinus*, *Betula*). During the Holocene Climatic Optimum (HCO), mixed broadleaf forests (deciduous *Quercus*, *Fraxinus excelsior* type, *Acer campestre* type, *Ulmus*, *Tilia*) reached their maximum extent, while cold-tolerant taxa declined. A cooling event at 8.2 ka BP led to reduced local precipitation and temperature, possibly driving tree cover reduction. Subsequent warming in the Holocene Thermal Maximum (HTM) coincided with *Abies* expansion, treeline rise, and subalpine forests establishment. In the mid-to-late Holocene, increased climatic variability resulted in fluctuations in tree cover, including *Abies* decline and *Fagus sylvatica* expansion. These changes could be probably linked to regional-local climatic influence and land use during the Copper and Bronze Ages, as documented in the area (2) and the Mediterranean region (3). Over the past ~3,000 years, a progressive tree cover reduction trend culminated with its local decline during the Medieval Climate Anomaly (MCA), marked by higher temperatures, positive Mediterranean Sea Surface Temperature (SST) anomalies, and a positive North Atlantic Oscillation (NAO) phase. The Little Ice Age (LIA) is characterized by increased anthropogenic indicators and wet meadow expansion. Despite overall resilience, montane and subalpine mixed forests show emerging signs of vulnerability. The findings emphasize the importance of conserving high-altitude peatlands and reinforcing the altitudinal ecological connectivity to support long-term ecosystem stability.

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4.11 = *Centaurea pumilio* L. Propagation and *ex situ* conservation

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Since its foundation, the Botanical Garden of the University of Salento has prioritized the *ex situ* conservation of endangered species native of the Salento region, particularly those listed on the National and Regional Red List. For many of these species, a long-term assessment of population size and threat factors have been observed to assign appropriate IUCN (International Union for Conservation of Nature) categories and to support scientifically justified initiative to reinforce population or reintroduction efforts (1).

Among these species is *Centaurea pumilio* L. (Asteraceae) also known as Cretan cornflower, classified a CR (Critically Endangered) on the Italian Red List and currently at great risk of extinction (2). The only known population in Italy is located in “Marina di San Giovanni”, in the municipality of Ugento (LE), where it was first recorded in 1996. *C. pumilio* is also present in other Eastern Mediterranean countries, including the Balkans, Turkey, Egypt, Palestine, Israel and Lybia, again with populations at risk; no protective measures or adequate conservation strategies have been implemented in any location, not even in Italy where the population continues to decline.

Here we report the results of generative propagation test carried at the Botanical Garden. Seeds were collected in June 2023, cleaned, and characterized by morphology (shape, color, size) and 1000 seeds weight; the accession was deposited in the seed bank. In October 2023, 100 seeds were taken to be sown in 7x7x18 cm PVC pots filled with a 1:1 mixture of peat and river sand. The pots were placed in an unheated tunnel. Germination reached 15% at 14th days after sowing and 85% by 30th day.

To assess possible declines in vitality over time, two further germination tests were conducted in January and March 2024. For each test 100 seeds per trial were distributed across three replicate Petri dishes moistened with deionized water. Dishes were placed in a phytotron under controlled conditions (16-hour photoperiod, 100 µE light intensity, and 22 °C constant temperature). Germination was monitored every two days, considering only seeds with fully emerged rootlets for calculating overall germinability and average three replicates.

A decline in seed vitality was registered over time. Germination rates decreased from 65% in January to 53% in March, with the average germination time increased from 102 days to 182 days. In both tests, germination began on 6th day post-sowing and reached approximately 50% between 20th and 22th days. January germination curve is completed within 30 days, while March curve is completed in 22 days

To increase the number of individuals eligible for *in situ* translocation and to support the recovery of the natural population, the generative propagation results achieved by the Botanical Garden complement the *in situ* conservation efforts of *C. pumilio*, as done in the past for other species (3).

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4.11 = Plant evolutionary history is largely underrepresented in conservation seed banks

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Considering the alarming prospect of at least two in five plant species facing extinction, it is urgent to identify unsecured phylogenetic branches within the plant Tree of Life and adopt appropriate conservation strategies. While conventional seed banking has the potential to safeguard a large part of world's flora, the scarcity of phylogenetically informed ex situ conservation programmes poses a challenge to effective plant conservation. Leveraging an extensive dataset of seed collections across 109 European seed banks embracing 121,418 seed lots, we found that among the 22,733 angiosperm plant species native to Europe, 8,383 (36.88%) are included in seed bank collections. Our study also reveals that current collections capture a phylogenetically diverse subset of the European flora. However, they safeguard between 43.29% and 66.40% of the maximum possible phylogenetic diversity, suggesting that specific major branches of the plant phylogeny in Europe remain unprotected.

To address this gap, we introduce a novel quasi-deterministic method to generate a list of unbanked species, prioritized by evolutionary significance. Although this approach can enhance the evolutionary quality of seed bank collections, biological, technical and practical constraints may limit conventional seed banking for some of these priority species.

We advocate for an enhanced coordination among conservation facilities and the integration of phylogenetic perspectives with advancements in ex situ conservation techniques beyond conventional seed banking, to effectively conserve plant evolutionary heritage.

4.11 = Conservation and Management of Annex I of Directive 92/43/CEE Habitats 3170* "Mediterranean Temporary Ponds" and 3130 "Oligotrophic to mesotrophic standing waters": Preliminary Results from the LIFE IMAGINE Project

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Many of the habitats listed in the Habitat Directive show an unfavorable conservation status, both in Europe and in Italy. To develop an effective conservation strategy, it is essential to experiment with different conservation and restoration practices to assess their potential effectiveness, with the ultimate goal of promoting and replicating those that prove to be most successful. This is the objective of the present study, carried out in the frame of the European project LIFE IMAGINE (LIFE19 IPE/IT/00015), which focuses on two habitats of European interest listed in Annex I of the Habitat Directive: 3130 and 3170*. Habitat 3170* (Mediterranean temporary ponds) consists of micro-mosaics of amphibious vegetation dominated by annuals and small geophytes, developing in late winter/spring in temporarily flooded, shallow, and waterlogged areas. Habitat 3130 includes amphibious vegetation—perennial or annual—growing in nutrient-poor, moist soils along the edges of stagnant inland waters (1,2). Our study focused on a valuable subtype (22.12 x 22.32) hosting the rare *Juncus bulbosus*, found in Umbria only in one site. Both habitats have a point-type distribution, are extremely localized and fragmented in Umbria, and lie at the climatic margin of their range, giving them high conservation importance. Our research focused on the ZSC IT5210020 “Boschi di Ferretto – Bagnolo”, where both habitats are present. We began by identifying the most significant pressures and threats affecting these habitats in the specific study area, and subsequently selected and mapped 66 intervention sites, equally distributed between habitats 3130 and 3170*(1). In each site, we identified specific pressures and/or threats, and planned and implemented targeted interventions. These interventions included removal of non-typical species; litter removal; shrub pruning; in some cases, the removal of alien species (such as the invasive moss *Campylopus introflexus*) and nitrophilous species (*Dittrichia viscosa*); and the translocation of germplasm of H-key species (in particular *Isoetes histrix* and *Juncus bulbosus*). All interventions were conducted manually—without the use of mechanical equipment—in order to avoid altering the delicate balance of these microhabitats (1). The work was carried out entirely by the staff of DSA3 (Unit of Botany). The work has been supported by targeted monitoring in the form of phytosociological surveys at the 66 intervention sites, separated into an *ex-ante* tranche, that is completed, and an *ex-post* tranche, which is still ongoing. Final results are not yet available. However, preliminary surveys indicate an increase in key species at the intervention sites. In particular, waterlogged soils (habitat 3170*) appear to benefit from the slight disturbance caused by our interventions. On the other hand, the effect of litter removal often appears to be temporary, especially in sites located near trees. Similarly, the manual removal of *Dittrichia viscosa* has shown limited effectiveness in areas where the species is widespread. In contrast, in areas where *Dittrichia viscosa* is less abundant, removal efforts have produced good results. The case of *Campylopus introflexus* is more complex. Due to its widespread presence and the high risk of regrowth from undetected patches, manual removal is particularly challenging. Nevertheless, in the areas where removal has been carried out, follow-up surveys have shown promising results, with no signs of the moss re-establishing itself so far. In conclusion, the results of this research may serve as a valuable foundation for developing more targeted management protocols for these fragile habitats. The aim is to offer useful guidance for landowners and institutions, with the ultimate goal of establishing formal agreements to implement regular management practices—such as thinning and shrub removal—that help maintain the low-intensity, ongoing disturbance essential for the long-term conservation of these habitats.

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4.11 = Does phylogenetic relatedness predict conservation status in Italian orchids?

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In an era of accelerating global biodiversity loss, understanding species' extinction risk within an evolutionary context is vital for effective conservation planning. Orchids, one of the most charismatic and ecologically specialized plant families, often face conservation challenges. Here, we examine the conservation status of 61 orchid species native to Italy through a phylogenetic lens. We map IUCN Red List categories onto a phylogeny constructed using the R package V.PhyloMaker, based on the GBOTB extended megatree of vascular plants, which incorporates the clade-level frameworks of Smith & Brown (2018) and Zanne et al. (2014)

Our analysis reveals that most of Italian orchids fall within the "Least Concern" category, suggesting relative ecological stability for many species. However, a subset of species classified as "Near Threatened" or "Endangered" forms distinct clusters within the phylogeny. This non-random distribution suggests that closely related species may share ecological traits, such as habitat specificity or pollinator dependence that increase vulnerability to environmental change, habitat fragmentation and over-collection.

Despite these insights, current research presents knowledge gaps, particularly in the spatial distribution and ecological requirements of Italian orchids. This highlights the need for comprehensive datasets combining conservation status, geographical range, and phylogenetic placement. Our ongoing work includes integrating distributional data into species profiles and visualizing them through maps and diagrams.

From a conservation standpoint, we advocate for the prioritization of phylogenetically distinct and vulnerable species in management plans. Incorporating evolutionary history into national conservation policies can improve resource allocation and help maintain both taxonomic and functional biodiversity. Furthermore, expanding this phylogenetic framework to encompass broader Mediterranean ecosystems could reveal deeper regional patterns of extinction risk and resilience.

Ultimately, our study underscores the power of phylogenetics in conservation biology—not only as a tool for understanding risk patterns but also as a strategic lens for anticipating and mitigating biodiversity loss in a changing world.

4.11 = Study of genetic variability of small populations of rare plant species: A case study of *Linnaea borealis* in Slovenia

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Glacial relict populations are isolated remnants of arctic-alpine species, resulting from the shifts of their distribution range during the glaciations. Recently, the conservation value of relict populations has been emphasized, since they are adapted to stressful ecological conditions, which may be important for future distribution range shifts due to climate changes, which occur rapidly in recent times. In Slovenia (Soteska by Nomenj) (Fig. 1) is the southeasternmost locality of twinflower in Europe, which grows in a low altitude of about 500 meters. The nearest sites are in Austrian Carinthia and Styria. Slovenian locality is therefore of a significant importance from biogeographic perspective. In this case, twinflower is a model species for the area that has been on the edge of permanent ice cover during the last glaciation. The genetic variability between two small, isolated populations of twinflower from the Eastern-Southeastern Alps (Slovenia and Austrian Carinthia) and three population from the core Boreal distributional range (Sweden, Scotland and Russia) were studied by means of nine microsatellite loci. The results revealed three main clusters, each indicating one population, while exemplars from the Boreal range are present in all three clusters (Fig. 2). Very low genetic variation within the two isolated Eastern-Southeastern Alpine populations and very high genetic variation between the two Eastern-Southeastern Alpine populations was found, despite being only 73 km apart. We conclude that the two isolated Eastern-Southeastern Alps populations are both glacial relict populations, based on the high genetic variability between them, probably due to consecutive bottlenecks and long-term isolation under specific environmental conditions. The isolated populations of twinflower in the Eastern-Southeastern Alps have not completely lost their capacity for sexual reproduction, so their survival “in situ” is still possible. However, glacial relicts, usually associated with vulnerable habitats deserve urgent attention involving conservation countermeasures especially with regard to the recent anthropogenically-driven environmental crisis.

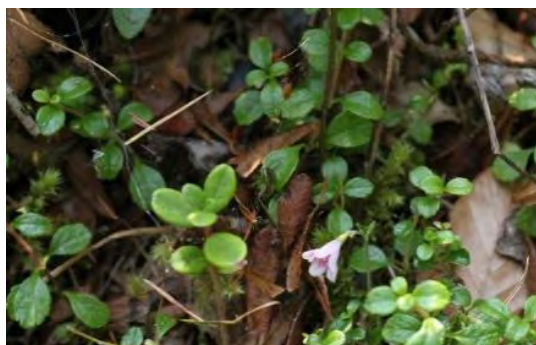


Fig. 1. *Linnaea borealis*

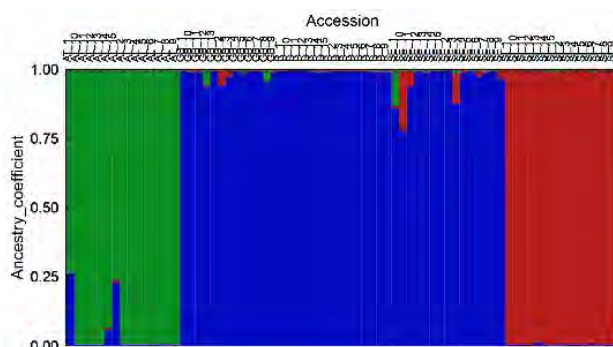


Fig. 2. Bar plot of Bayesian analysis results

1) N. Pipenbaher, M. Kaligarič, S. Škornik, D. Ivajnsič, T. Ternjak, M. Šiško (2026) Acta botanica Croatica: an international journal of botany, 85

2) T. Wraber (1963) *Linnaea borealis* L., planta rediviva slovenske flore [*Linnaea borealis* L., planta rediviva of the Slovenian flora]. Prirodoslovni muzej, Ljubljana

5.3 = Fungal Contamination in Jan Wildens' Paintings: Isolation, Characterization, and Biocide Screening

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Fungal colonization is the greatest danger to canvas paintings, as it can trigger a chain of enormous biodeterioration processes that destroy cultural heritage objects' aesthetic value and structural integrity. The selection of appropriate biocidal treatments remains a challenging and crucial step in conservation processes, especially due to the variability of fungal resistance and the potential vulnerability of artworks to chemical substances.

Our investigation studied two oil-on-canvas paintings of the Flemish artist Jan Wildens (Antwerp, 1586 – Antwerp, 1653) to assess the active fungal infection and effective control strategies.

Diagnostic analysis was preceded by ultraviolet fluorescence and visual examination to detect fungal growth if present. These samples were then drawn from infected areas and subjected to a polyphasic identification technique that combined morphological analysis with molecular techniques (e.g., ITS sequencing), enabling accurate classification of the obtained fungal strains from the paintings.

Fungal strains were then used in a series of viability and inhibition tests to evaluate the effectiveness of a commercial biocide, Biotin R1 + R2, at a 3% concentration. The biocide was both in vitro and applied to canvas strips directly to simulate real application conditions.

Our results demonstrate this integrated strategy's effectiveness in identifying viable biodeteriogen fungal strains and testing them for sensitivity to specific biocides. This allows the most effective conservation treatments to be selected with fewer risks to the original materials.

This study emphasizes the need for targeted microbiological tests in heritage conservation and advocates for the implementation of targeted, evidence-based restoration practices.

5.3 = Fungal contamination in a university archive: an aeromycological approach

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Aeromycology investigates airborne fungal particles (PBAPs), which are essential for evaluating indoor air quality (IAQ), especially in environments affected by moisture damage. High humidity, water infiltration, and poor ventilation lead to fungal growth on substrates such as paper, wood, plaster, and rubber, resulting in biodeterioration and health risks (1). Fungal exposure has been linked to a variety of health issues, including Sick Building Syndrome (SBS), allergic responses, and infectious diseases (2). This study assesses fungal contamination in a university archive subjected to flooding from heavy rainfall and degradation of rubber liners in mobile shelving systems. Three sampling points were selected, and a total of 18 plates were exposed to 3,600 liters of air. The air was sampled using the active Surface Air System (SAS), with 200 liters of air aspirated per plate. The samples were cultured on three different media (Rosa Bengal Agar, Sabouraud Dextrose Agar, and Malt Extract Agar). The airborne fungal load in the archive reached 299.72 CFU/m³, which corresponds to intermediate contamination according to INAIL guidelines, but exceeds the recommended 150–200 CFU/m³ threshold set by the Italian Ministry of Culture for archival environments. More than 90% of the isolates belonged to the genera *Aspergillus*, *Penicillium*, and *Cladosporium*, with *Aspergillus creber* Jurjević, S.W. Peterson & B.W. Horn 2012 as the most prevalent species. Seven species of *Penicillium*, including *P. chrysogenum* Thom 1910 and *P. brevicompactum* Dierckx 1901, were found, both associated with rubber degradation. Additionally, *Mucorales*, listed as high-priority pathogens by the WHO, were identified. All strains will be preserved in the COLD-UNIGE microbial collection, a node of the MIRRI network, for future taxonomic and functional studies. The Fungal Susceptibility Index (FSI), as developed by Di Piazza et al. (2024) (3), was applied to evaluate the degradation risk to various materials and the pathogenic risk for humans.

1) J. D. Smith, "Fungal Contamination in Indoor Environments," *Indoor Air Quality Journal*, vol. 14, no. 2, pp. 35-42, 2015

2) M. T. Stevens et al., "Health Effects of Fungal Exposure," *Environmental Toxicology*, vol. 11, no. 3, pp. 23-30, 2020

3) Di Piazza, L., et al., "Fungal Susceptibility Index for Material Degradation Risk Assessment," Abstract presented at the 2023 International Mycology Congress (IMC), Maastricht, Netherlands, 2024

5.5 = Towards a selection of plant species for Italian rain gardens: role, characteristics and taxa

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Climate change is a major concern threatening mankind and natural ecosystems, growing as the rising temperatures alter the global energy and hydrological cycles worldwide. As seen in recent years, heat waves, droughts, heavy precipitation, and windstorms, critically affect European cities. Nature Based Solutions (NbS) effectively address climate-related threats, providing at the same time multiple benefits known as ecosystem services (ESs), or, in the most recent definition, “nature contributions to people” (NCP). Sustainable stormwater management techniques arose in the last decades in many countries around the world, under different and at different scales. Urban flooding mitigation is the focus of practices and programs such as Low Impact Development and Green Stormwater Infrastructures (USA), Water-sensitive Urban Design (Australia), the Sponge City Initiative (China), and Sustainable Urban Drainage Systems (UK and Europe). From rain gardens and tree trenches to constructed wetlands, these green infrastructures provide a wide range of well-recognized benefits. In addition to managing stormwater, they support biodiversity, improve the urban microclimate, and capture harmful pollutants. Vegetation plays a central role in each of these systems, being crucial to both the extent and quality of their positive impacts. Indeed, plants’ appropriateness has consequences not only in qualitative and quantitative stormwater management, but also in the development of low-maintenance management strategies suited to the challenging conditions of the built environment, helping to enhance local biodiversity, increase the diversity of plant and animal species, and reduce the risk of biological invasions. Species’ selection and arrangement criteria, therefore, are analyzed and presented with their implications (1,2). Specifically, the current study reviewed both international scientific and grey literature concerning the selection of plant species for use in rain gardens, to present an updated resource for researchers and designers. First, the main characteristics individuated by scientific literature, involving rooting system and aerial parts, are presented to highlight the possible guidelines for new selection of *taxa*. Both water quantity and quality management are considered (1,2,3). Then, the most frequently used herbaceous and woody species are listed and grouped by geographical and ecological range, also considering their potential invasiveness if used in the Italian context. Finally, the Italian indigenous species found in literature are analyzed, highlighting strengths and criticalities, referring to climatic stresses and other ecological aspects. A review of selection criteria and potentially suitable other species are then introduced, to guide further research on the topic and advance the application of rain gardens in Italy.

1) Greksa, A., Ljubojević, M., Blagojević, B., 2024. The Value of Vegetation in Nature-Based Solutions: Roles, Challenges, and Utilization in Managing Different Environmental and Climate-Related Problems. *Sustainability* 16, 3273

2) Corduan, D., Kühn, N., 2024. Planting for the Urban Rain—Vegetation in Urban Bioretention Systems for Stormwater Management under Temperate Climate Conditions—A Systematic Review. *Sustainability* 16, 8861

3) Técher, D., Berthier, E., 2023. Supporting evidences for vegetation-enhanced stormwater infiltration in bioretention systems: a comprehensive review. *Environ Sci Pollut Res* 30, 19705–19724

5.6 = Chasing a Ghost Plant: *Scabiosa trenta*, Julius Kugy, and the Cultural Heritage of Exploring the Julian Alps

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The story of *Scabiosa trenta* Hacq., first described in 1782 by Balthasar Hacquet and never detected again in nature, exemplifies how taxonomic ambiguity can fuel scientific ambition and inspire cultural imagination. Indeed, believed to be endemic to Slovenia's Trenta Valley, this elusive species became the focus of a decades-long botanical mystery. Central to this narrative is the figure of Julius Kugy, whose youthful encounter with the mystery of *S. trenta* in 1877 catalyzed a profound engagement with the Julian Alps, marking the beginning of a lifelong dedication to alpine exploration and botanical inquiry. Kugy's search for *S. trenta*—conducted with the support of local guides and in dialogue with contemporary botanists—epitomizes the intersection of early alpine science and romantic naturalism (1). Though ultimately unfruitful in locating the species, Kugy's expeditions laid critical groundwork for botanical surveying in the Julian Alps. His evolving perspective, from taxonomic pursuit to geographic and cultural exploration, reflects a broader shift in how mountain landscapes were scientifically and symbolically perceived in the late 19th century. His close collaboration with Albert Bois de Chesne led to the establishment of the Juliana Alpine Botanical Garden in Trenta, a lasting institution of alpine biodiversity and public education. It was the botanist Anton Kerner von Marilaun who later resolved the taxonomic puzzle, identifying *S. trenta* as a high-altitude ecotype of *Cephalaria leucantha* (L.) Roem. & Schult. —a finding that demonstrated the importance of herbarium revision in clarifying species identity (2). Yet, far from being a scientific footnote, the tale of *S. trenta* persisted as a cultural metaphor for the unattainable, embedded in literature, folklore, and regional identity. This contribution aims to explore how the pursuit of *S. trenta* represents more than a botanical case study: it is a story of how taxonomy, personal exploration, and the alpine environment coalesced into a powerful legacy. Through Kugy's journeys, the Julian Alps emerged not only as a site of scientific curiosity but also as a space where cultural heritage was written into the landscape—where the boundaries between science, storytelling, and identity became strongly entangled (3).



Fig. 1. Original drawing of *S. trenta* belonging to the protologue



Fig. 2. *C. leucantha* growing in the Alpine Botanical Garden of Trenta (Slovenia)

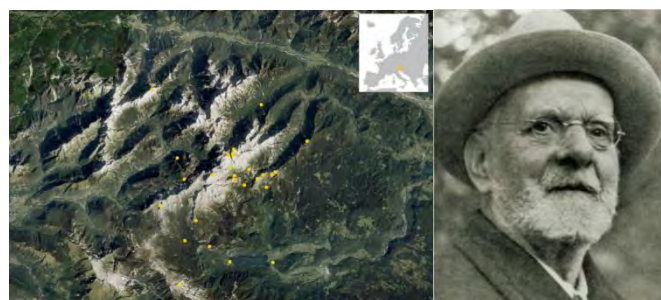


Fig. 3a-b. Julius Kugy and a map showing the mountain summits he explored to find *S. trenta* (yellow dots)

1) Rozman, K. 1987. The British, Kugy, and Western Slovenia. *The Alpine Journal* 92: 63-71

2) Kerner von Marilaun, A. 1893. *Scabiosa Trenta* Hacquet. *Österreichische Botanische Zeitschrift* 43: 113–117. Retrieved from <https://www.biodiversitylibrary.org/page/28826838>

3) Conix, S. 2019. Taxonomy and conservation science: interdependent and value-laden. *History and Philosophy of the Life Sciences* 41(2): 15. <https://doi.org/10.1007/s40656-019-0252-3>

5.6 = Magnetic and chemical biomonitoring with lichens and vascular plants for the preservation of cultural heritage: a case study at two museums in a megacity (Buenos Aires, Argentina)

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Lichen transplants are commonly employed in air biomonitoring due to their effectiveness in accumulating important levels of particulate matter (PM) and potentially toxic elements (PTEs). Their usage in the field of cultural heritage offers a natural and inexpensive technique for assessing pollutants, that may settle and subsequently harm artworks. Biomonitoring surveys were recently conducted in cultural heritage sites to outline the source, pattern of diffusion, and concentration of PM and PTEs in close proximity of the sites. Magnetic and chemical biomonitoring took place at the Museo Nacional de Bellas Artes (MNBA) and at the Museo Histórico Nacional (MHN), two museums situated along highly traveled roadways in Buenos Aires, Argentina. Lichen of the species *Evernia prunastri* was selected as biomonitor and transplanted from November 2022 to March 2023 in both museums of Buenos Aires. A mixed sampling approach has been established, incorporating four outdoor exposure spots at MNBA and three at MHN, together with three indoor locations in both museums. Together with the lichen deployment, plant leaves (*Jacaranda mimosifolia* at the first site and *Fraxinus americana* at the second one) were sampled at the beginning and the end of the lichen exposure. Lichen exposure aimed at assessing the diffusion of vehicular emissions from outside to inside the museum's halls. The leaf analysis aimed at evaluating their suitability for providing preventive conservation services. Lichen and leaf samples were analysed for magnetic susceptibility by mass (χ , m³/kg) and hysteresis properties, i.e. the coercive force (B_c , T), the saturation remanent magnetization by mass (M_{rs} , Am²/kg) and the saturation magnetization by mass (M_s , Am²/kg). Afterwards, samples were analyzed to quantify the content of PTEs related to anthropogenic activities, i.e. Al, Ba, Cr, Cu, Fe, Mn, Sb, Zn. The magnetic properties of leaves and lichen transplants highlighted a significant accumulation of magnetic and coarse particles, sourced mainly from brake abrasion, in the area facing the museum at both sites (Fig. 1); the indoor transplants, on the other hand, did not deviate significantly from the unexposed lichen. Both leaves and outdoor lichen transplants accumulated relevant concentrations of PTEs (Ba, Cd, Cr, Cu, Fe, Mn, Sb and Zn), showing vehicular emissions as the main pollution source (especially for Sb and Cd, mainly derived from brakes, Fig. 2). On the contrary, lichens did not significantly bioaccumulate PTEs inside the museums. In this experiment, lichen was verified to be a viable biomonitor of airborne PM and PTEs, indicating clearly that PTEs accumulation was insignificant indoors, although the exterior accumulation of the same elements was significant. Moreover, *J. mimosifolia* showed an impressive ability to accumulate the target pollutants on their leaves, proving also a better performance than *F. americana* leaves. This outcome suggested that *J. mimosifolia* provides efficient ecosystem service retaining high burden of PM and PTEs.

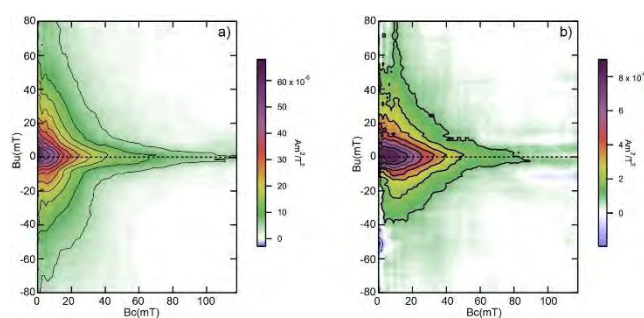


Fig. 1. FORC diagrams for selected leaf (a) and lichen (b) at MNBA

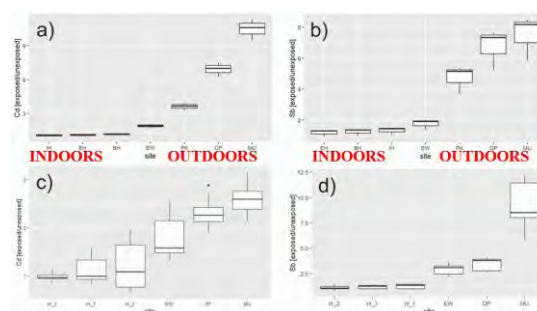


Fig. 2. Lichen exposed-to-unexposed ratios for Cd and at each site of MNBA (a, b) and MHN (c, d)

5.6 = Contribution to the ethnobotanical knowledge in communities of Northern Maremma (Tuscany): the case study of Tirli village

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Ethnobotany is a fundamental tool for understanding the historical and cultural relationships between human communities and the plants world. Today, it plays an important role in conserving biodiversity and promoting local flora. This study explores the traditional plant knowledge within the Tirli community, nestled in the heart of Maremma (GR, Tuscany), a representative example of regional cultural heritage. The research is an integral part of the B.E.T.A. project, included in the 2014–2020 Rural Development Programme (PSR) of the Tuscany Region for the regeneration of local communities. Using 45 semi-structured interviews, we gathered insights from 31 respondents (12 men, 19 women; average age 61), focusing on the culinary and medicinal uses of wild plants. Over half reported personally using of wild species, with 42% being lifelong residents—strong evidence of a deep-rooted connection to their land. We documented 70 wild plant species, mainly used for food and traditional medicine. Among the most cited were *Clinopodium nepeta* (L.) Kuntze subsp. *nepeta* with Use Report (URs) = 16, *Anethum foeniculum* L. and *Urtica dioica* L. (URs = 12 each), all showing the highest Cultural Importance (CI up to 0.533). This index combines both the frequency of mention, and the diversity of uses attributed to each species, thereby capturing the depth and breadth of ethnobotanical knowledge associated with them. The Relative Importance (RIs) index and Cultural value (CvE) for ethno-species highlighted the central role of *Urtica dioica*, *Clinopodium nepeta* subsp. *nepeta*, and *Malva sylvestris* L., confirming their key function in transmitting ethnobotanical knowledge. The Fidelity Level (FLs) per species analysis revealed a remarkable 100% match between species and reported use categories for over 60 taxa, underscoring the cultural specificity of these uses. Additionally, the Number of Uses (NUs) per species showcased the versatility of several plants, linked to multiple use categories by different informants. The chord diagram showed the predominance of food-related links, but also a significant representation of medicinal knowledge. Although food uses dominate, many of these plants are well-documented in ethnobotanical literature for their medicinal properties, suggesting future research opportunities: plants used primarily as food in Tirli may hold untapped therapeutic potential. These culturally versatile species enrich local knowledge systems and offer key resources for integrated strategies aimed at preserving both biological and cultural heritage. Overall, this study highlights the richness and vitality of Tirli's ethnobotanical knowledge, showing how tradition can become a powerful ally in sustainability and sustainable land management.

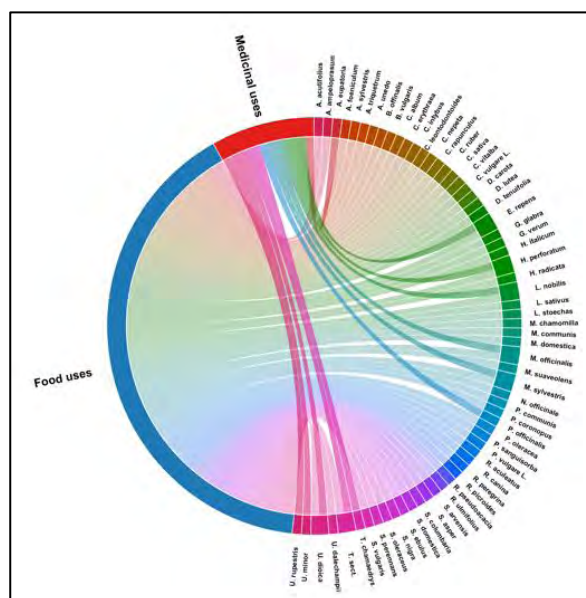


Fig. 1. Chord diagram showing the relationships between wild plant species and use categories (food and medicinal) in the community of Tirli

- 1) Motti, R., Paura, B., Cozzolino, A., & Falco, B. D. (2022). Edible flowers used in some countries of the Mediterranean basin: An ethnobotanical overview. *Plants*, 11(23), 3272
- 2) Pentassuglia, M., Lombardi, T., Bambi, G., Ventura, I., D'Ambrosio, B., Bertacchi, A., & Pistelli, L. (2025). A Floristic Survey of Wild Edible Plants in Tuscan Maremma, Italy. *Plants*, 14(6), 976
- 3) Sulaiman, N. (2025). Botanical Ethnoknowledge Index: a new quantitative assessment method for cross-cultural analysis. *Journal of Ethnobiology and Ethnomedicine*, 21(1), 20

5.7 = Cultural ecosystem services of urban street trees: functional traits

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Urban street trees provide a wide array of benefits, or Ecosystem services, to citizens. These benefits may be enhanced selecting specific species traits and they can be tangible or intangible, as most Cultural Ecosystem Services (CESs). CESs in the urban context include benefits for the health and wellbeing of citizens, but also aesthetic, spiritual and cultural values and street trees contribute to providing these benefits (1, 2). However, although the benefits included within CESs are universally recognized as commodities, their categorization, quantification, estimation and comparability are still problematic.

We performed a literature review (about 60 papers up to now) on CESs provided by urban street trees, in relation to their traits. Specifically, as a first step, we selected the CESs that urban street trees can provide (i.e., Recreation and mental and physical health, Tourism, Aesthetic appreciation and inspiration for culture art and design, Spiritual experience and sense of place). Secondly, we performed a literature review to link species traits to the selected ecosystem services. As a final step, we developed a framework, based on information retrieved in the literature [see (2)], which shows how specific traits are more commonly associated with CESs. This approach has several positive aspects, for instance, traits are properties that are intrinsic to the species and, as such, they are objective features that do not change among different cities (as a consequence, it is easier to compare results and analyses) (3). Traits can be easily selected to increase the provision of Ecosystem services and reduce potential disservices (2) however, CESs seem to vary among cultural regions and within the same region. This is especially true for the aesthetic appreciation (i.e., a preference for tall or short trees, for the shape of the tree, etc.), while other CESs seem to be less variable. For instance, shading trees are preferred while exercising or walking.

Our study suggest that people have preferences for specific tree attributes, however, according to research there are also other factors that pedestrians prefer as, for instance, a combination of green elements along streets, such as trees, shrubs and flowers and also residents seem to prefer a diversity of tree species.

1) J. Mullaney, T. Lucke, S.J. Trueman (2015) *Landsc. Urban Plan.*, 134, 157-166

2) V. Savo, L. D'Amato, F. Bartoli, I. Zappitelli, G. Caneva (2025) *Ecosyst. Serv.*, 71, 101690

3) J. Goodness, E. Andersson, P.M. Anderson, T. Elmqvist (2016) *Ecol. Indic.*, 70, 597-605

6.1 = Invasive potential of *Cotula coronopifolia* L. in Italian coastal wetlands

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Invasive alien species threaten ecosystems, biodiversity and human well-being, generating high economic costs. Due to its cultural and commercial centrality, the European Union is particularly exposed to the introduction of alien species. *Cotula coronopifolia* L. (Asteraceae) (Fig. 1) is native of Southern Africa and it is reported as introduced in other five continents, including Europe. This species colonizes both brackish and freshwater wetland environments and it is reported as invasive in Italy (1), with confirmed occurrences in five regions, including Campania. The presence of this species in Campania has been documented within the salt marsh ecosystems of the Variconi SAC (IT8010028), and later observed in the nearby Soglitelle Nature Reserve (2). This study contribute to evaluate the invasiveness of *C. coronopifolia* within habitat types listed in Annex I of the Council Directive 92/43/EEC by: a) evaluating the germination response of *C. coronopifolia* under six levels of salinity concentration; b) comparing germination performances of native species living in the same environments and typical of salt marshes habitats (Fig. 2); c) analyse changes in *C. coronopifolia* distribution in the Variconi SAC. Within the Variconi Nature Reserve, the same trails of the earlier study (2) were resampled to record the presence of *C. coronopifolia*. For the germination experiments, seeds collected from multiple individuals were placed in Petri dishes with filter paper moistened with chloride solutions simulating increasing salinity levels. Following established protocol (3), dishes were kept in a controlled chamber for 20 days, with germination monitored every two days and solutions regularly replaced to maintain constant salinity. Updating of the new distribution data revealed that *C. coronopifolia* has further expanded, failing to invade only in areas where salinity levels are too high. The germination tests confirmed that *C. coronopifolia* has a good tolerance to salinity, but only up to concentrations of 7.5 g/L Cl, slightly higher than other native species, except that of *Salicornia fruticosa* (L.) L., a well-known halophyte highly tolerant to salt. The findings confirmed the species capacity to establish itself and rapidly expand within brackish environments, representing a potential threat to native plant species and local vegetation assemblages.



Fig. 1. *Cotula coronopifolia* individuals



Fig. 2. *Salicornia fruticosa* stands invaded by *Cotula coronopifolia*

1) Galasso G, et al. (2024). A second update to the checklist of the vascular flora alien to Italy. Plant Biosystems 158(2): 297– 340

2) Marfella, L, et al., 2023. Distribution of the invasive alien species *Cotula coronopifolia* L. (Asteraceae) relating to water halinity and sodicity in the Variconi wetland (Campania, southern Italy)." Hydrobiologia 850: 1653-1668

3) Strumia, S. et al., 2020. Seed germination and seedling roots traits of four species living on Mediterranean coastal cliffs. Plant Biosystems 154(6), 990–999

6.1 = First results of the monitoring of alien species in the Capo Peloro area (Messina, Sicily)

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In the north-eastern corner of Sicily, north of Messina, there is a promontory that narrows and ends with the tip of Torre Faro, known as Capo Peloro. It is an extension of the Peloritani mountain range that slopes gently towards the sea with a hilly system, mainly consisting of a Quaternary deposit of sand and gravel of various shapes and sizes (fossil dunes) called "Ghiaie di Messina". Geographically, it extends mainly in a west-east direction, even if in the first stretch it has a south-west, north-east trend (1); the climate is decidedly subtropical with discrete rainfall (between 800 and 700 mm per year) and an average annual temperature of around 20 °C. This has favoured the spread of alien species originating from tropical and subtropical climates, rather than temperate species which are found more in hilly areas and on slopes with a northern exposure. Particularly, among the alien species of temperate environments, the dominant ones are *Robinia pseudoacacia* L. and *Ailanthus altissima* (Mill.) Swingle, deliberately introduced as consolidants of landslide slopes, and which cover the highest parts of the hills and the slopes exposed to the north and the more humid valley floor where they compete with the remains of woods dominated by *Quercus pubescens* Willd. subsp. *pubescens*, *Quercus suber* L. and *Pinus pinea* L. Among the alien species of sub-tropical and tropical origin we find well distributed those belonging to the genera *Agave* (5 species: *A. americana* L. subsp. *americana*, *A. angustifolia* Haw. subsp. *angustifolia*, *A. attenuata* Salm-Dyck subsp. *attenuata*, *A. salmiana* Otto ex Salm-Dyck subsp. *ferox* (K.Koch) Hochstätter, *A. sisalana* Perrine), *Aloë* (4 species: *A. arborescens* Mill., *A. maculata* All. subsp. *maculata*, *A. vera* L., *A. ×caesia* Salm-Dyck), *Asparagus* (3 species: *A. aethiopicus* L., *A. falcatus* L., *A. setaceus* (Kunth) Jessop), *Opuntia* (3 species: *O. ficus-indica* (L.) Mill., *O. stricta* (Haw.) Haw., *O. tuna* (L.) Mill.), *Yucca* (3 species: *Y. aloifolia* L., *Y. gigantea* Lem., *Y. gloriosa* L.) and *Washingtonia* (2 species: *W. filifera* (Gloner ex Kerch., Burv., Pynaert, Rodigas & Hull) de Bary, *W. robusta* H.Wendl.). The most represented families are: Poaceae (8 genera and 8 species: *Arundo donax* L. *Cenchrus setaceus* (Forssk.) Morrone, *Dactyloctenium aegyptium* (L.) Willd., *Digitaria ciliaris* (Retz.) Koeler, *Echinochloa colona* (L.) Link subsp. *colona*, *Megathyrsus maximus* (Jacq.) B.K.Simon & S.W.L.Jacobs, *Saccharum biflorum* Forssk., *Setaria adhaerens* (Forssk.) Chiov.), Asparagaceae (3 genera and 11 species: *Asparagus aethiopicus*, *A. falcatus*, *A. setaceus*, *Agave americana* subsp. *americana*, *A. angustifolia* subsp. *angustifolia*, *A. attenuata* subsp. *attenuata*, *A. salmiana* subsp. *ferox*, *A. sisalana*, *Yucca aloifolia*, *Y. gigantea*, *Y. gloriosa*), and Arecaceae (3 genera and 5 species: *Butia capitata* (Mart.) Beccari, *Phoenix canariensis* H.Wildpret, *P. dactylifera* L., *Washingtonia filifera*, *W. robusta*). The concentration of alien species is particularly high in the northern outskirts of the city of Messina, along the highly anthropized coastal strip and particularly within the Riserva Naturale Orientata "Laguna di Capo Peloro", due to its very favorable soil and climate characteristics (2). The ways in which these alien species have spread are multiple, both in terms of methods and opportunities: most come out of gardens and private homes, while a small part entered the area following the transit routes from the city of Messina. The diffusive strategy can be implemented with the production of edible fruits and fertile seeds as in the genera *Asparagus*, *Butia*, *Lantana*, *Passiflora*, *Phoenix*, *Yucca*, *Washingtonia* and the species *Ficus rubiginosa* Desf. ex Vent., *Schinus terebinthifolia* Raddi; dispersion of fertile seeds through wind or various vectors such as in *Cenchrus setaceus*, *Boerhavia coccinea* Mill., *Catharanthus roseus* (L.) G.Don, *Setaria adhaerens*; mixed diffusion through fertile seeds and asexually as in the genus *Opuntia*, and the species *Austrocylindropuntia subulata* (Muehlenpf.) Backeb., *Ailanthus altissima*, *Cyperus alternifolius* L. subsp. *flabelliformis* Kük., *Robinia pseudoacacia*, *Saccharum biflorum*; spread by asexual means as in the genus *Agave* and the species *Yucca gigantea*. In conclusion, the presence of these alien species does not seem to have an impact on the area except in anthropized environments, particularly they do not seem to cause disturbances or reductions in the relict population of *Tricholaena teneriffae* (L.f.) Link present in the study area (1).

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6.1 = Reliable data to tackle Alien Plant Invasions: the PREVALIEN Geo-Database for modelling, early detection and management in Protected Areas

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As part of the PRIN 2022 PREVALIEN project - aimed at enhancing knowledge on PREvention and early detection of the invasive ALIEN plants of (European) Union concern in the Italian protected areas - coordinated by the University of Sassari, the first work package (WP1) focused on developing the relational PREVALIEN geo-database (GDB). This GDB is designed to integrate, harmonize, and make accessible data on Invasive Alien Plants of Union Concern (IAPUC) occurring in Italian protected areas. The PREVALIEN-GDB was built using the PostgreSQL RDBMS with the PostGIS extension to support spatial data management and analysis. This GDB was developed following international standards, such as those of the European Alien Species Information Network (EASIN). Each of the 41 IAPUC is identified by a unique code based on Regulation (EU) No. 1143/2014 and its updates, ensuring consistency across data layers. The core data were primarily extracted from the official Pest Risk Analyses and Risk Assessments available through the EU Communication, the Information Resource Centre for Administrations, Businesses and Citizens (CIRCABC), and the European and Mediterranean Plant Protection Organization (EPPO) repositories. These resources provided detailed, structured, and expert-reviewed data on species introduction history, pathways, ecological impacts, and available management strategies. This information was further enriched by integrating data from international and national databases [e.g., EASIN, TRY(1), GLONAF(2), CABI, Portal to the Flora of Italy (3)], scientific literature, local floras, herbaria, targeted field surveys, and citizen science platforms such as iNaturalist. The PREVALIEN-GDB organizes this information across 18 data tables, which reflect key dimensions of biological invasions. These include taxonomy and phylogeny, introduction history, habitat preferences, and species distribution at global, European, national, and regional scales. Other data tables focus on species traits such as growth form, specific leaf area, dispersal modes, and other ecological features. This structure allows for multi-dimensional analyses, as well as the development of species models and early warning tools for a better management of biological invasions. The PREVALIEN-GDB provides the necessary infrastructure to improve the knowledge about IAPUC in protected areas, supporting robust analyses of ecological, landscape, and socio-economic drivers of IAPUC spread. Furthermore, the database will serve as a baseline for the development of a broader monitoring framework, extendable to the European scale.

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- 3) Portal to the Flora of Italy. Available at <http://dryades.units.it/floritaly>

6.4 = Park age shapes ecosystem services in urban lawns

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Urban green spaces are essential elements of modern cities, providing several ecosystem services (1). To understand how soil characteristics and different layers of biodiversity (vegetation, arthropods and microbes) have an impact on ecosystem services, we sampled and analyzed soils from urban lawns in five major Italian cities along a latitudinal gradient (Turin, Milan, Florence, Rome, Naples), in parallel to adjacent areas with a lower anthropic disturbance as control (remnant grassland).

The national-scale sampling campaign was conducted from summer to fall 2023 and included data collection across different biodiversity layers from soil sampling for 16S/ITS2 amplicon sequencing (meta-barcoding) to enzymatic activities essays, physico-chemical analyses, vegetation survey and determination of the soil biological quality index based on microarthropods (QBS-ar). By integrating these data with remote sensing information (NDVI) we estimated the main ecosystems services provided by urban lawns, including soil biological quality, ability to control pests and establish plant symbioses, organic matter decomposition, carbon sequestration, nutrients availability, soil stability and porosity, and net primary productivity. First, ecosystems functions (EF) were estimated using as proxies measured variables including physico-chemical parameters, soil enzymes activities and from the total amount of relative abundances of selected fungal taxa belonging to a given functional group according to the FungalTraits database. These functions were standardized between 0 and 1, according to Wang and colleagues (2) to define ecosystems functions. These functions were integrated into the ecosystem services mentioned above (mean of the functions for each service) and integrated into the ecosystem multifunctionality (EMS) index by averaging each ecosystem service. Similarly, multidiversity and multidissimilarity was estimated by normalizing diversity and dissimilarity index of each group (prokaryotes, fungi and plants) and by averaging normalized values into the multidiversity and multidissimilarity indices. Ecosystem services were then compared across park types using ANOVA followed by Tukey's post-hoc test for normally-distributed data or the Kruskal-Wallis test with Dunn's post-hoc test for non-normal data ($p < 0.05$). Structural Equation Modelling (SEM) was performed with the 'lavaan' R package (3) to study relationships between soil ecosystem services and environmental features.

Our results indicate that the multifunctionality (*i.e.* the average of ecosystem services measured) of urban park lawns is lower than that of less disturbed areas (control). Moreover, the number of ecosystem services that reach at least 75% functionality is higher for historical lawns than recent ones. These data suggest that, in the long run, park lawns tend to increase ecosystem services. Only the decomposition of organic matter and soil porosity were significantly lower in urban parks, while no differences were observed in terms of nutrient availability or plant symbiotic associations. Notably, the number of fungal pathogens was higher in recent lawns than in historical ones. Interestingly, microbial communities profiling across the latitudinal gradient revealed low dissimilarity in urban lawns, while increased values occurred for control sites. This suggests a homogenization of soil microbiota in urban sites while high variability occurred in less disturbed sites. The use of a structural equation model revealed that both park age and percentage of clay have a positive direct effect on multifunctionality in urban lawns. The city and mean annual precipitation also have a weak indirect influence on multifunctionality.

Overall, these data improve our understanding of ecosystem services provided by biodiversity in urban green spaces, paving the way for designing healthier and more sustainable cities.

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6.4 = The real taste of our waste: deciphering the role of compost microbial diversity in supporting tomato growth and drought resistance

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Over the past half-century, the need to feed a rapidly growing global population has led to intensive agricultural practices, resulting in widespread soil degradation and biodiversity loss (1). This, in turn, has reduced the availability of microbial strains that can positively interact with the plants. Among the sustainable strategies to address this issue, compost stands out as a promising solution, as it supports a circular economy and has long been used in agriculture for its organic matter and nutrient content. Moreover, it has been observed that the addition of compost can have some effects on the interaction between plants and beneficial microorganisms; therefore, compost could be considered as a good candidate for maintaining and improving the soil microbial biodiversity (2). However, the composition and functional role of compost microbiota in enriching soil communities and promoting plant health require further research. For this reason, a characterization of the microorganisms present in the compost is needed to further broaden the possibilities of tailoring specific solutions for its application.

This study, conducted in collaboration with S.E.S.A S.p.A., demonstrated that compost-associated microbiota positively influences plant growth in various ways. To investigate whether compost treatment induces changes in the plant-associated bacterial community, tomatoes were cultivated using compost, sterile compost, and a chemical fertilizer; a metagenomic analysis was then performed on samples from the bulk soil, the rhizosphere and the roots. The results showed that bacterial communities in the presence of compost were significantly different (in terms of beta diversity) from those in plants treated with sterile compost or chemical fertilizer. Moreover, the rhizosphere of compost-treated plants exhibited greater bacterial richness and evenness, indicating higher alpha diversity within this compartment that is crucial for plant-microbe interactions.

Many of the enriched bacterial taxa belonged to families known for plant growth-promoting traits, such as *Bacillaceae* and *Pseudomonadaceae*. Additionally, the presence of bacterial taxa associated with plant stress responses prompted an investigation into compost microbiota performance under drought conditions. Tomato plants subjected to severe water deficiency and treated with compost maintained higher levels of photosynthetic efficiency compared to those treated with sterilized compost or a chemical fertilizer. These plants also showed upregulation of genes involved in nutrient uptake and phytohormone signaling, which support overall plant health.

These findings underscore the potential benefits of compost in enhancing plant-associated microbial communities. This research contributes to the advancement of sustainable agricultural practices and the promotion of soil ecosystem health, in line with the EU's 2030 sustainable development goals.

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7.1 = Diversity of Medicinal Species and Compounds in the Hymenochaetaceae Family

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The Hymenochaetaceae family (Agaricomycetes, Basidiomycota) includes a wide diversity of wood-inhabiting macrofungi, with several species historically used in traditional medicine and increasingly recognized for their biotechnological potential [1]. Recent studies report at least 124 medicinally relevant species across 27 genera, producing approximately 500 mycochemicals, including terpenoids, styrylpyrones, steroids, phenolics, and polysaccharides. Notably, genera such as *Inonotus*, *Phellinus*, *Sanghuangporus*, and *Fuscoporia* have yielded novel bioactive metabolites like gilvsins, hispolon, inonotins, phellibarins, and baumin, many of which are structurally unique in the fungal kingdom. Documented pharmacological activities exceed 100 types, with antioxidant, antitumor, antidiabetic, immunomodulatory, anti-inflammatory, and antimicrobial effects most frequently reported. Specific compounds such as fuscoporic acid and inotolactones exhibit cytotoxic, antiviral, and enzyme-inhibitory properties with potential applications in cancer therapy and metabolic disorders [2]. Molecular phylogenetic analyses, using maximum likelihood methods, revealed that these medicinal species are distributed among 27 well-supported monophyletic clades. Importantly, 20 species are generic types, anchoring the taxonomic framework of their respective genera. The majority of medicinal Hymenochaetaceae are native to Asia, particularly China, reflecting both natural diversity and intensive ethnopharmacological use. However, the high frequency of taxonomic misidentifications in biomedical literature underlines the need for accurate molecular characterization to ensure reproducibility and specificity in pharmacological applications. These findings collectively highlight the Hymenochaetaceae as a prolific and underexploited source of novel bioactive compounds and reinforce the necessity of integrating taxonomy, phylogeny, and mycochemistry for future biomedical exploitation.

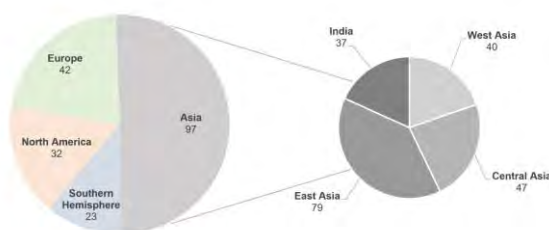


Fig. 3: Geographic distribution of the medicinal species of Hymenochaetaceae. Numbers refer to the number of species

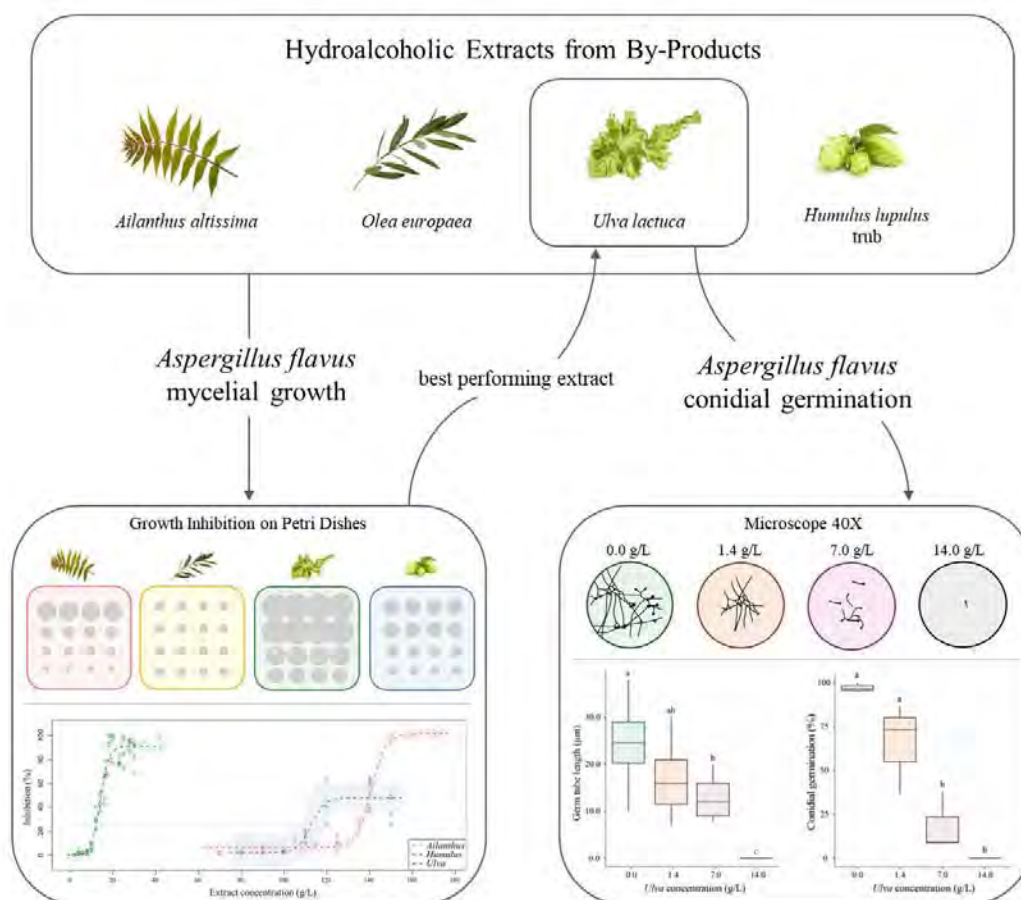
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7.1 = *In vitro* antifungal effect of plant-based and *Ulva* sp. extracts on *Aspergillus flavus* growth and conidia germination

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The accumulation of mycotoxins in cereals, and particularly in maize, is one of the most hazardous consequences of climate change in agriculture. Therefore, the development of effective strategies to mitigate this threat is urgently needed. Plant and seaweed extracts represent promising sources of phytochemicals against fungal plant pathogens. This study evaluated the *in vitro* antifungal activity of hydroalcoholic extracts derived from plant waste and seaweed against *Aspergillus flavus*, a fungal pathogen of maize. Different concentrations of extracts from *Ailanthus altissima* leaves, *Humulus lupulus* trub, *Olea europaea* leaves and *Ulva* sp. thallus were tested by agar-well diffusion assay against pathogen mycelial growth. All extracts except *O. europaea* inhibited fungal growth, *Ulva* sp. being the most effective extract with $EC_{50} = 14.2$ mg/mL, followed by *A. altissima* ($EC_{50} = 140.7$ mg/mL) and *H. lupulus* trub ($EC_{50} = 113.7$ mg/mL), the latter being unable to completely inhibit the mycelial growth even at the highest concentration tested. Conidial germination was affected by increasing concentrations of *Ulva* sp. extract. The polyphenolic profiles of the extracts revealed they were mainly enriched in glycosylated flavonols and hydroxycinnamic acids, whereas *Ulva* sp. showed a pronounced content in rosmarinic and *p*-hydroxybenzoic acid. *Ulva* sp. extract contained 24% uronic acid and 31% sulfate groups, characteristics of ulvan polysaccharides. Rosmarinic acid, *p*-hydroxybenzoic acid and ulvan pure compounds were individually tested *in vitro* against *A. flavus*, confirming their involvement in the inhibition of pathogen mycelial growth. Results indicated the promising biopesticide potential of *Ulva* sp. extract against *A. flavus*, although its application under field conditions requires further investigation.



7.1 = NanoBios: biocompatible nanosystems for enhanced delivery of plant biostimulants

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As global food demand rises and environmental concerns intensify, the search for more sustainable agricultural practices becomes increasingly urgent. A promising approach to reduce crop production dependence on chemical fertilizers is represented by plant biostimulants, substances or microorganisms that enhance plant nutrition and growth independently of their nutrient content (1). Biostimulants have been shown to improve the overall plant performance and stress tolerance, acting even at very low concentrations. Nevertheless, their application in the field can be limited by their chemical or physical status, namely their susceptibility to environmental degradation or volatility. Nanotechnology offers an innovative strategy to address this issue, specifically through the nanoencapsulation of the bioactive compounds, which may enhance not only the biostimulant stability and resistance to environmental degradation, but also the efficiency of their delivery to plants (2). The present work aims to develop, in collaboration with the company ILSA S.p.A., novel nanoencapsulated eco-friendly biostimulant fertilizers and biopesticides, collectively called NanoBios. Two classes of nanosystems are being employed in the nanoencapsulation, Metal Organic Frameworks (MOFs) and Hydrogen-bonded Organic Frameworks (HOFs), which have been selected for their high porosity, tunability and biocompatibility. Preliminary synthesis trials, conducted using MOFs nanosystems, achieved the successful encapsulation of the bioactive molecules, as confirmed by the characterization of the NanoBios via transmission and scanning electron microscopy, as well as X-ray diffraction and mass spectrometry. NanoBios are currently being tested in *Arabidopsis thaliana* cell suspension cultures (3), to rule out potential cytotoxic effects of the nanosystems and evaluate their possible internalization within plant cells. Viability assays showed the absence of cytotoxic effects of empty HOFs, and fluorescently labelled nanosystems are being developed to analyse NanoBios cellular uptake via laser scanning fluorescence microscopy. Moreover, aequorin-based Ca^{2+} measurements in *Arabidopsis* seedlings and suspension-cultured cells are ongoing, to uncover the potential calcium-mediated sensing of NanoBios. This study contributes to the establishment of a solid analytical platform to screen future formulations and sets the groundwork for subsequent investigations into the plant growth-promoting/priming activity of NanoBios *in vivo*.

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7.1 = Exploring the antiviral potential of small metabolites from mass cultures of *Staurosirella pinnata* and *Cylindrotheca closterium*

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Diatoms, a diverse group of microalgae, produce a wide range of bioactive metabolites in response to environmental stimuli (1). These compounds play vital roles in stress adaptation, intercellular communication, and defense, and have attracted growing interest for their potential pharmaceutical applications, particularly in the treatment of cancer (2), metabolic diseases (3), and microbial infections caused by viruses. Beyond their ecological roles, these compounds represent a promising source in biomedical research, particularly in drug discovery.

This study aimed to evaluate the antiviral potential of low-molecular-weight (<3 kDa) metabolite fractions obtained from two benthic diatom species: *Staurosirella pinnata* (Ehrenberg) D.M. Williams & Round, isolated from brackish sediments of the Cabras Lagoon (Sardinia), and *Cylindrotheca closterium* (Ehrenberg) Reimann & J.C. Lewin, isolated from coastal marine samples in Latium. Both strains were mass cultivated in 8-liter cylindrical photobioreactors under standardized conditions: 25 °C, 80 µmol photons m⁻² s⁻¹ irradiance, and a 12:12 h light/dark cycle. After reaching stationary phase, the biomass was harvested, lyophilized, and subjected to methanolic extraction followed by size-based fractionation via ultrafiltration to isolate the <3 kDa metabolite pool.

These fractions were assessed for their antiviral activity against three RNA viruses with different host specificities and replication strategies: feline coronavirus (FCoV, a surrogate model for human coronaviruses), Sendai virus (SeV, a murine parainfluenza virus used as a model for respiratory infections), and Influenza A virus (WSN strain, H1N1 subtype). Extracts were applied to infected Vero and MDCK cell lines at a final concentration of 2.0 mg/mL, and viral replication was analyzed by means of cellular assays.

Results revealed strong inhibitory effects on viral replication: *S. pinnata* extract fractions reduced FCoV replication by up to 97%, while both diatom extracts inhibited SeV replication by up to 85%. *C. closterium* also showed significant activity against WSN, achieving up to 81% inhibition. Cytomorphological observations of FCoV-infected cells treated with *S. pinnata* extract further demonstrated preserved cellular integrity, suggesting not only antiviral but also cytoprotective activity. No significant cytotoxicity was observed in uninfected cells treated with the extracts, supporting their potential safety.

Overall, this study highlights the capacity of diatom-derived small metabolites to interfere with viral replication across different viral models, underscoring their promise as a source of novel antiviral agents.

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7.2 = Enhancing *ex situ* cultivation of Mediterranean Fucales in an ecological restoration perspective: species-specific responses of *Gongolaria barbata* and *Ericaria crinita* seedlings to algal extracts

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Mediterranean brown algal forests, dominated by *Cystoseira sensu lato* species, are undergoing widespread decline due to the cumulative effects of anthropogenic pressure and climate-related stressors. Restoration efforts increasingly rely on *ex situ* cultivation and outplanting of seedlings, yet early developmental stages often suffer from low survival and growth rates. This study investigated the potential of algal-derived extracts to enhance the seedling development and survival in two canopy-forming species, *Gongolaria barbata* and *Ericaria crinita*.

We tested extracts from a cyanobacterium (*Trichormus variabilis*), two microalgae (*Desmodesmus* sp. and *Cylindrotheca closterium*), and a commercial macroalgal formulation (AlgatronCifo®) at varying concentrations under controlled mesocosm conditions.

Seedling performance was significantly influenced by extract type, and target species identity. Notably, a low-concentration *Desmodesmus* sp. extract (0.07 mg mL⁻¹) improved survival and growth, whereas *T. variabilis* exerted an inhibitory effect on *G. barbata*. AlgatronCifo® did not outperform *Desmodesmus* sp. extract in promoting seedling development.

These findings suggest that specific extracts from green microalgae could improve protocols for the early stages of restoration, offering a scalable tool for rehabilitating degraded marine forests. However, the results underscore the importance of species-specific optimization and the need for in situ validation of biostimulant-based restoration approaches.

7.2 = Efficacy of lignin nanocapsules for delivering neem oil and capsaicin against *Plutella xylostella*

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Over the past 50-60 years, pesticides have been largely used in agriculture, causing adverse effects on the ecosystem and humans. For this reason, antiparasites administered in the form of finely dispersed particles, the so-called nanopesticides, have recently gained attention.

In this work, we report on the design, preparation, and physico-chemical characterization of nanosystems based on kraft lignin for the transport of neem oil and capsaicin as insect repellents. The methodology followed was aimed at establishing a protocol for the scaling up, which can also ensure that the obtained dispersions are stable in water media, from where they can be administered in a safe and easy way (e.g. foliar spray).

Lignin was initially dispersed in a KOH solution to obtain a concentration of 5% w/w. After neem oil addition (4.5% v/v), the resulting dispersed aggregates were sonicated to reduce their size. To increase the insect repellency effect, capsaicin was added to the oil phase. In-depth physico-chemical characterization by Dynamic Light Scattering, electron microscopy, and Small Angle X-ray Scattering showed that all formulations showed dimensions in the range of 300–400 nm with moderate polydispersity and globular shape, which tended to be more elongated in the case of lower starting pH. Negative zeta potential values were measured in all the samples, thus ensuring good stability by electrostatic repulsion.

From preliminary results against *Plutella xylostella*, neem oil and capsaicin carried by lignin submicron capsules efficiently exert their action when tested on *Eruca sativa* plantlets and this effect persisted for seven days, avoiding their eating.

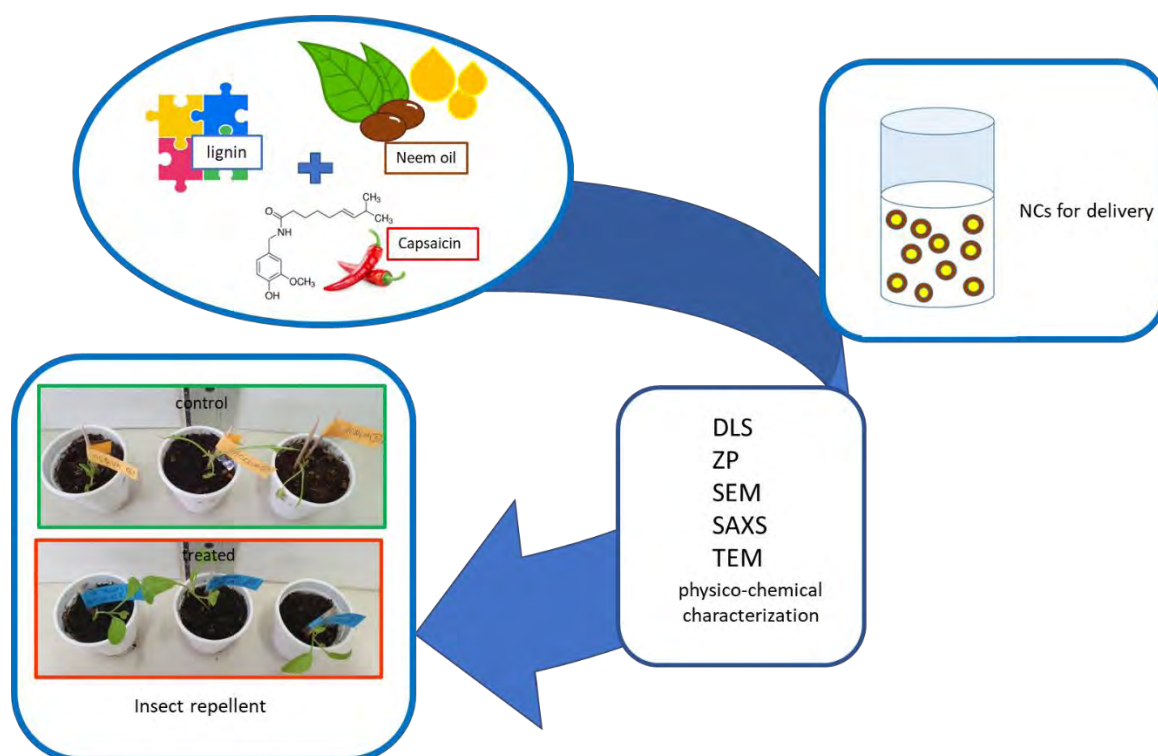


Fig. 1. Novel strategy based on lignin nanocapsules to deliver capsaicin and neem oil against *Plutella xylostella*

1) S. Falsini, T. Nieri, A. Papini, M. C. Salvatici, A. Abou-Hassan, C. Gonnelli and S. Ristori Efficiency of lignin nanocapsules for delivering neem oil and capsaicin against pest insects: insights into the system *Eruca sativa*– *Plutella xylostella*, Environ. Sci.: Nano, Environ. Sci.: Nano, 2025,12, 1922-1929

7.3 = Hard endosperm tissue in edible seeds: the case of *Coffea arabica* and *Phoenix dactylifera*

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In certain seeds, including some of the endospermic legumes, the major storage product is deposited in the cell walls as hemicellulose. In species such as fenugreek, ivory nut, coffee and date, the hemicelluloses, largely galactomannans, may be deposited in large quantities in the peculiar and thick cell walls of the storage organ that characterize these types of seed, such as the endosperm tissue. This tissue is extremely hard because of the chemical structure of cell wall polysaccharides (1). This morphoanatomical feature connects coffee and date seed, but very few data were reported about the similarities of their endosperm tissue. This study aims to describe the morphoanatomical characteristic of endosperm tissues in both *Coffea arabica* and *Phoenix dactylifera*, in order to highlight differences and similarities that could be interesting for applications in the food industry.

Portion of seeds of *C. arabica* and *P. dactylifera* were fixed and washed in distilled water, then cut by a cryostat at -15°C, obtaining transversal seed sections of 12 µm of thickness. Sections were treated with TBO solution for metachromatic stain to highlight the main anatomical seed characteristics and observed by an optical microscope (Leica DM2500). Other sections were observed by a SEM (Hitachi TM3030plus).

The metachromatic stain underlines the main different seed tissues, such as the inner integument and the endosperm in both coffee and date. In particular, date endosperm tissue is characterized by a massive, thick-walled endosperm, rich in pure mannan and by cells rich in protein and lipid bodies (2). Coffee endosperm is very similar to date endosperm, but cell walls are rich in galactomannans and arabinogalactans (3) and the cellular lumen is twice as big as date cells. In contrast, cell walls in coffee are less thick and wavier than date cell walls. Primary pit fields were particularly evident in date cells.

These characteristics in common of endosperm tissue suggest similar behavior under intense industrial treatments such as roasting processes and the consequent induced chemical changes. Some authors suggested date seed as a caffeine-free coffee surrogate and several products containing coffee and date seed blends have been already launched on the market.

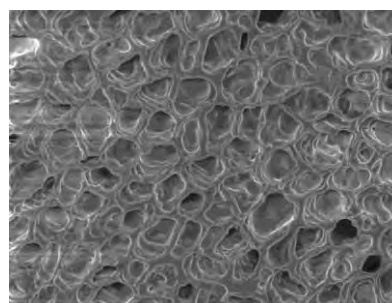
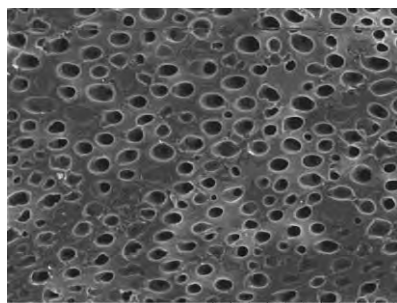


Fig. 1. Endosperm tissue of *P. dactylifera* (SEM, bar: 200 µm)

Fig. 2. Endosperm tissue of *C. arabica* (SEM, bar: 200 µm)

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7.3 = Treat it responsibly: how soil microbial metabolism responds to cover crops and *Trichoderma asperellum* as biostimulant

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Soil depletion and fatigue due to the decrease in organic matter, degradation and consequent reduction in metabolic activity is one of the consequences of prolonged exploitation in agricultural systems. From the past to the present, a sustainable agronomic practice for controlling weeds, breaking pest and disease cycles, and improving soil fertility has been the use of cover crops in rotation with cash crops [1]. Besides, microorganisms (bacteria and fungi) have been more recently applied as crop enhancers in “bioaugmentation”. For example, several species in the fungal genus *Trichoderma* Pers. are applied in agriculture to favour the overall plant growth (and roots especially), as well as to stimulate the induced systemic resistance and suppressing conditions towards certain plant pathogens [2].

The general aim of this work was to evaluate the variation in metabolic activity in soils from experimental cultivations of the *Capsicum annuum* L. (Solanaceae) “Voghera pepper” ecotype and in relation to differently combined treatments relying on cover crops and biostimulation by *Trichoderma asperellum* Samuels, Lieckf. & Nirenberg. More specific questions are: how do these two different treatments interact with each other? What effects may they have on the soil metabolism? How do these results contribute to suggesting good practices in soil management?

Soil was sampled at three timepoints: before cover crop sowing (November 2023), soon after the cover crop flowering (April 2024) and at bell pepper ripening time (October 2024). Soil samples were furtherly added with a suspension of the same *T. asperellum* strain in a mixed experimental design, then incubated at 25 °C for 15 days. Diluted soil suspensions were subsequently analysed by EcoPlates™ (Biolog) methodology, *i.e.* 96-well microplates containing 31 different carbon sources (in triplicate). The evolution of metabolic profiles by time for each treatment was surveyed for 21 days by periodical spectrophotometric check.

The results showed that soil metabolic activity was enhanced in samples from cover crop parcels, regardless of whether the *T. asperellum* biostimulant was used or not. On the other hand, *T. asperellum* application significantly changed metabolic activity in the last timepoint (bell pepper ripening) in the parcels where cover crops had not been applied, *i.e.* in the most fatiguing and depleting condition for the field soil. The different utilization of individual carbon sources as by EcoPlates™ analysis in fact highlights the effect of *T. asperellum* in restoring metabolic activity and microbial competence. The cover crop treatment thus resulted in an overall enhancement of soil metabolic activity, whereas *T. asperellum* contributed to support the fruiting effort and restore the soil balance in absence of cover crops.

This work is part of the Proof of Concept (PoC) Project “SAPEVO” (*Integrated Strategies of Biostimulation Applied to Voghera Bell Pepper*) within NODES, funded by MUR on *fondi M4C2 -Investimento 1.5 Avviso “Ecosistemi dell’Innovazione”*, within PNRR funded by the European Union – NextGenerationEU (Grant agreement Cod. n. ECS00000036) Spoke 6 CUP F17G22000190007.

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7.3 = Study of the Combined Effects of Biostimulants and Reduced Nitrogen Fertilization in Tomato Plant Development and Fruit Quality

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Nitrogen (N) is one of the main nutrients required by plants to ensure healthy and productive growth, participating in numerous key biochemical processes. Nevertheless, the excessive use of nitrogen fertilisers, adopted to maximise agricultural productivity, has over time led to negative impacts on the environment, including soil and groundwater contamination.

In the context of global climate change, which already threatens the productivity of many staple crops for the agri-food economy, it is certainly crucial to develop resilient and sustainable cropping systems; therefore, traditional fertilisation practices could be insufficient to meet the new production and ecological challenges.

An innovative and promising solution is biostimulants, substances or microorganisms capable of improving plant nutritional efficiency, tolerance to abiotic stresses and crop quality (1). Among the most studied biostimulants are protein hydrolysates, seaweed extracts and beneficial microorganisms such as nitrogen-fixing endophytic bacteria. These products, according to scientific evidence, are able to stimulate soil microbial activity, promote nutrient uptake and support plant growth even under conditions of limited N supply (2).

Tomato (*Solanum lycopersicum* L.) is a globally widespread horticultural crop, particularly sensitive to nitrogen availability, that can significantly affect both yield and crop quality. In greenhouse production, in particular, the amount of N required can vary considerably depending on the stage of plant development and the specific growing conditions. Furthermore, it has been observed that an increase in nitrogen above a certain threshold does not necessarily lead to an improvement in productivity but may reduce fruit quality and increase costs and environmental impact (3).

This study aims to analyse the efficacy of three different categories of biostimulants - protein hydrolysates, algae extract and an endophytic nitrogen-fixing bacterium - applied to tomato plants subjected to different levels of nitrogen fertilisation. The objective is to evaluate their impact on plant development, fruit quality and nitrogen use efficiency, with the aim of identifying strategies that reconcile high productivity and high fruit quality and reduced environmental impact.

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7.3 = Bio-sustainable fungicides from by-products of the beer supply chain

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This research work is part of a larger project, called BIO-RESYPE (PRIN 2022 - CUP D53C24004480006), born out of a collaboration between the Universities of Ferrara, Turin and Cattolica del Sacro Cuore in response to growing concerns about the environmental impact of traditional agricultural practices and increasing consumer demand for safer food (1).

Hop by-products represent a sustainable source of valuable biomolecules that can be obtained through circular economy strategies utilising eco-friendly extractions and separations (2).

Unfermented and fermented spent hops were hydrodistilled to obtain essential oils and aromatic water. The chemical characterization of essential oils showed α -humulene, β -caryophyllene and β -myrcene as main compounds. The essential oil of the fermented spent hops also evidenced the presence of ethyl esters of fatty acids. The chemical characterization of the aromatic water from fermented spent hops is in progress.

An *in vitro* experiment has been conducted to test the possible capacity of these extracts to reduce the growth of *Fusarium graminearum* and, more importantly, the production of deoxynivalenol (DON), a mycotoxin toxic to both humans and animals.

Petri dishes with artificial medium were inoculated with 1 isolate of the mycotoxigenic fungus and incubated at 4 different temperatures (15, 20, 25, and 30°C) in the absence and in the presence of three hop extracts (essential oils and aromatic water).

The results showed good ability to contain fungal growth with greater reductions at low temperatures (73 to 77%) and less at high temperatures (33 to 65%). On the other hand, with regard to DON containment, reductions were greater at 30°C (50 to 87%) and fair at other temperatures (24 to 66%).

Based on the results obtained, a field trial will be scheduled to test how weather conditions and cultural practices may affect the effectiveness of these naturally occurring substances.

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7.5. = *Daphne laureola* roots: a promising source of bioactive compounds with antiangiogenic and antioxidant activity

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The genus *Daphne* (Thymelaeaceae) comprises over 90 species distributed across Asia, Europe, and North Africa. These plants have long been employed in ethnomedicine throughout China, Tibet, Korea, and the Middle East to treat a variety of health conditions (1).

Numerous studies have revealed the remarkable biological potential of *Daphne* species, which are rich in pharmacologically active compounds, including coumarins, flavonoids, lignans, steroids, and terpenes. Many *Daphne* species exhibit a broad range of biological effects, such as antibacterial, antifungal, antioxidant, analgesic, anti-inflammatory, antiviral, and haemostatic activities (2).

Daphne laureola L. (spurge laurel) is a Mediterranean evergreen shrub traditionally used in South Italy as an ethnoveterinary remedy. The present research focuses on the study of the chloroform-methanol extract of *D. laureola* roots with the aim of investigating its phytochemical composition, antioxidant, and antiangiogenic activity. A preliminary phytochemical screening of this extract, obtained after static maceration using solvents of increasing polarity, evidenced a high content of total polyphenols and flavonoids (146.41 ± 4.58 mg gallic acid equivalents/g DW and 35.77 ± 2.44 mg catechin equivalents/g DW, respectively).

The antiangiogenic activity was evaluated using two *in vivo* models: the chick chorioallantoic membrane (CAM) and zebrafish embryo assays (3). Results demonstrated that the extract induced a dose-dependent inhibition of neovascularization in both experimental models, supporting its capacity to modulate angiogenic processes *in vivo*. Thus, the extract was subjected to Biotage® flash chromatography followed by RP-HPLC to obtain some pure flavonoids, coumarins, and lignans that were tested in the same assays. Among them, buddlenol D, rutarensin, and neochamaejasmin B emerged as the most active compounds, due to the strong antiangiogenic effects suggesting their key role in the overall activity of the chloroform-methanol extract.

Subsequently, the antioxidant potential of the extract and its isolated compounds was evaluated through DPPH, ABTS, and FRAP assays. Results showed that the extract exhibits strong radical scavenging activities (113.00 ± 2.65 mg TE/g for DPPH, 238.65 ± 1.82 mg TE/g for ABTS) and a good reducing capacity (106.27 ± 5.12 mg TE/g for FRAP). Among the isolated compounds, buddlenol D evidenced the best antioxidant activity in all the assays.

In conclusion, these findings highlight *D. laureola* roots as a valuable source of bioactive compounds with antiangiogenic and antioxidant properties. The identification of bioactive constituents supports the therapeutic potential of *D. laureola* root for future pharmacological investigations aimed at developing novel agents to counteract pathological conditions supported by uncontrolled angiogenesis and oxidative stress.

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7.5 = Phytochemical and Biological Characterization of Extracts from *Allium ursinum*: A Study on a Traditionally Used Plant

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Allium ursinum L., commonly known as wild garlic, is a wild edible plant traditionally used across Europe as a spring tonic, appreciated for its digestive, diuretic, and purifying properties. Ethnobotanical records document the use of the whole plant – leaves, bulbs, and flowers – in decoctions, fermented preparations, and topical applications, reflecting a holistic approach to its medicinal use. Despite its long-standing presence in folk medicine, comprehensive pharmacological evaluation remains limited. In this study, *A. ursinum* specimens were collected in the forests of Rosello (Chieti, Italy) and analysed for their phytochemical profile and biological activities. Aqueous and hydroalcoholic extracts (EtOH 50% and 70%) were obtained from whole dried plant material and assessed for total phenolic and flavonoid content, antioxidant capacity, and enzyme-inhibitory activity. The 50% hydroalcoholic extract exhibited a good polyphenolic profile (TPC: 23.19 ± 1.04 mg GAE/g_{dm}) and demonstrated effective radical-scavenging and reducing capacity. It also demonstrated notable inhibitory activity against selected enzymes, including acetylcholinesterase, tyrosinase, and glucosidase. These effects appear to be at least partially related to its phenolic content. The extracts were tested for allelopathic activity, brine shrimp lethality, *Daphnia magna* heartbeat effects, and cell viability on C2C12 and WiDr cell lines, showing low ecotoxicity and extract-specific biological responses. In C2C12 cell-based assays, the 50% hydroalcoholic extract showed the most consistent cytoprotective effect under oxidative stress (H₂O₂ 750 μ M), compared to the aqueous and 70% ethanol extracts. Based on this integrated chemical and biological profile, the 50% hydroalcoholic extract was selected for further studies, including gene expression analysis on *ex vivo* mouse colon tissues. Markers related to inflammation and oxidative stress – specifically COX-2, IL-6, and iNOS – were investigated to explore the extract’s potential effects on inflammatory pathways. By combining traditional uses with experimental evidence at both the phytochemical and biological levels, this study contributes to validating the ethnomedical relevance of *A. ursinum* and highlights its promise as a source of functional bioactive compounds.

7.5. = Phenolic content, antioxidant properties, and brine shrimp toxicity of leaf extracts from *Erucastrum virgatum* subsp. *virgatum* (Brassicaceae) endemic to Sicily (Italy): a comparative study of different collection times

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Erucastrum virgatum C.Presl subsp. *virgatum* (Brassicaceae) is an endemic perennial herbaceous plant, typical of only 3 regions of Southern Italy: Sicily, Calabria, and Basilicata (1). Rather rare, in Sicily it is found only in the province of Messina, distributed exclusively on the limestone promontories of the Ionian coast (Capo Scaletta, Capo Ali, Capo S. Alessio, Capo Taormina) and of the Tyrrhenian coast (Capo Tindari, Capo Milazzo). It can also be found on the island of Ustica.

Ethnobotanical studies report the use of leaves and flowering shoots of this species in traditional Sicilian cuisine (2,3). Despite its food use, there are no studies in the literature regarding the phytochemical composition or evaluation of biological properties. Therefore, in continuation of our investigations into species included in the Brassicaceae family growing wild in Sicily, it seemed interesting to undertake a study on *E. virgatum* subsp. *virgatum*.

The present work aimed at evaluating the antioxidant potential of leaf extracts of *E. virgatum* subsp. *virgatum* collected in the locality of Scaletta Zanclea (Messina) at different times during the year: January (*Ev-Jan*), March (*Ev-Mar*), June (*Ev-Jun*), and November (*Ev-Nov*). The leaves were lyophilized and subjected to extraction in an ultrasonic bath at 50°C using an EtOH-H₂O mixture (70:30 v/v) as green solvent.

The extracts were investigated for their phenolic content and antioxidant properties. The quantitative determination of total phenolics and flavonoids was attained by spectrophotometric methods. The antioxidant properties were determined by different *in vitro* methods: the primary antioxidant activity was evaluated by the 2,2-diphenyl-1-picrylhydrazyl (DPPH) and reducing power assays, while the secondary antioxidant properties were estimated by the ferrous ion chelating activity assay. Furthermore, *Artemia salina* lethality bioassay was performed for preliminary toxicity assessment.

The total phenolic content was found to be higher in *Ev-Jan* extract (65.15 ± 1.78 mg GAE/g extract), and decreased in the following order: *Ev-Jan* > *Ev-Mar* > *Ev-Jun* > *Ev-Nov*. The same trend was observed for the flavonoid content (52.07 ± 1.75 mg QE/g extract). All leaf extracts of *E. virgatum* subsp. *virgatum* showed mild to moderate primary antioxidant properties compared to the standard BHT, with *Ev-Jun* displaying the best DPPH radical scavenging activity ($IC_{50} = 1.54 \pm 0.02$ mg/mL) and reducing power (32.06 ± 0.13 mg ASE/g extract). For both tests, the activity of the extracts decreased as follows: BHT > *Ev-Jun* > *Ev-Jan* > *Ev-Mar* > *Ev-Nov*. In the ferrous ion chelating activity assay all extracts exhibited good activity compared to the standard EDTA, which was found to be higher for *Ev-Nov* ($IC_{50} = 0.25 \pm 0.01$ mg/mL), exhibiting the lowest content of both phenolics and flavonoids, and decreased as follows: EDTA > *Ev-Nov* > *Ev-Mar* > *Ev-Jan* > *Ev-Jun*. The results of the antioxidant tests indicate that the extracts from the leaves of *E. virgatum* subsp. *virgatum* at different collection times possess antioxidant properties based on diverse mechanisms. Finally, the results of *A. salina* lethality bioassay showed the absence of toxicity against brine shrimp larvae for all the extracts.

Our findings contribute to enrich the knowledge of the biological properties of *E. virgatum* subsp. *virgatum*, so far unexplored, indicating this edible species that grows spontaneously in Sicily as novel resource of natural antioxidants.

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7.5 = Preliminary analysis on exhausted *Myrtus communis* L. berries

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The study of exhausted berries of *Myrtus communis* L., a by-product of myrtle liqueur production, is part of the broader effort to recover and valorize agri-food residues. Despite the substantial production of myrtle liqueur at the national level, the exhausted berries resulting from the extraction process have received limited scientific attention. However, these materials may still retain noteworthy amounts of bioactive compounds with potential nutraceutical and cosmetic applications.

In this preliminary investigation, an initial objective was to identify the most suitable extraction solvent to maximize the recovery of the residual bioactive constituents within the exhausted berries. This step aimed to optimize the extraction efficiency of the natural phytocomplex, while also comparing the extraction capacity of various methodologies. The resulting extracts were then subjected to a series of analytical evaluations, including the quantification of total phenolic compounds, flavonoids, anthocyanins, and flavan-3-ols. Additionally, the antiradical activity was assessed using the DPPH (2,2-diphenyl-1-picrylhydrazyl) radical scavenging assay, while the antioxidant activity was measured using the FRAP assay. To further investigate the qualitative differences of the composition, the phenolic profiles of the extracts were analyzed by high-performance liquid chromatography with diode-array detection (HPLC-DAD).

Preliminary findings revealed the presence of considerable residual concentrations of bioactive molecules, indicating that the berries, even after undergoing the industrial extraction process for liqueur production, still preserve relevant levels of these compounds. These results highlight the potential of exhausted *M. communis* berries as a valuable source of natural bioactives, suitable for further exploitation.

Future work will involve more comprehensive chemical characterization of the extracts and *in vitro* studies to assess the biological properties of the recovered compounds. These investigations are intended to explore possible therapeutic, pharmaceutical, and cosmetic applications, contributing to the development of sustainable strategies for by-product utilization.

7.5 = Research and Open Science at the Ghirardi Botanic Garden (Toscolano Maderno - Brescia, Italy): a multi-level study on *Santolina chamaecyparissus* L. (Asteraceae)

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A multi-level survey was conducted on *Santolina chamaecyparissus* L. (Asteraceae), cultivated at the Ghirardi Botanic Garden (Toscolano Maderno, Brescia, Italy) of the University of Milan. This work is part of two wider ongoing projects entitled *Ghirardi Botanic Garden, factory of molecules...work in progress* (active from 2021) and *Green Circuit, journey in and out of the botanic garden* (started in 2024); both projects are intended to preserve, enhance and spread knowledge on the plant heritage hosted at the Ghirardi Botanic Garden.

The multidisciplinary study combined three sequential phases: *i*) micromorphological and histochemical, to describe the structures responsible for the productivity of secondary metabolites, and to define the chemical nature of the secretory products, by means of Light Microscopy (LM) and Scanning Electron Microscopy (SEM); *ii*) phytochemical, to characterize the essential oil (EO) profile obtained from the aerial parts at blooming in two consecutive years (2021 and 2022) by Clevenger-type hydrodistillation coupled with Gas Chromatography-Mass Spectrometry (GC-MS); *iii*) biological, with a literature research to define the potential biological activity of the most abundant EO compounds.

The micromorphological survey on *S. chamaecyparissus* was performed on the vegetative and reproductive organs. We observed uniseriate non-glandular trichomes, very variable in length, and two different types of secretory structures: external glandular trichomes (biseriate and ten-celled) and internal ducts. The former exclusively produced an oleoresin, while the latter a resin rich in flavonoids. The analysis of the EO composition revealed comparable quali-quantitative profiles over the two consecutive years, with 38 and 40 components, in 2021 and 2022, respectively, representing 98.75% and 96.60% of the total. The dominant compound classes were oxygenated monoterpenes (57.68%, 58.55%) and monoterpenes hydrocarbons (18.91%, 19.41%), followed by oxygenated sesquiterpenes (16.27%, 14.88%). The main compounds were camphor (20.57%, 21.43%), artemisia ketone (18.43%, 16.59%) and α -bisabolol (11.14%, 9.07%). Finally, based on literature data on EOs with camphor and artemisia ketone as ones of the main compounds (1,2), antimicrobial, antioxidant, and anti-inflammatory properties were hypothesized for the EO analyzed herein.

In the light of an *Open Science* policy, this information was combined in the creation of novel interpretative apparatuses on the target-species at the Ghirardi Botanic Garden. In this way the results of the scientific research are made accessible to the visitors, giving the opportunity to discover the plant heritage under an unusual perspective, starting from the macroscopic features towards the microscopic secrets.

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7.5 = Chemical constituents and cytotoxic activity on colon cancer cells of *Cynara cardunculus* subsp. *cardunculus*

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Cynara cardunculus subsp. *cardunculus* L. [*Cynara cardunculus* L. var. *sylvestris* (Lam.) Fiori] (wild artichoke, *C. sylvestris*) is a spiny species related to the cultivated artichokes [*C. scolymus* L., *C. cardunculus* var. *scolymus* (L.) Fiori, *C. cardunculus* subsp. *scolymus* (L.) Hegi]. It is present in the Mediterranean with different ecotypes and other subspecies including *C. cardunculus* subsp. *flavescens* Wiklund, *C. cardunculus* subsp. *zingaroensis* (Raimondo & Domina) Raimondo & Domina, all closely related. *C. sylvestris* stands out as the primary wild taxon prized for its culinary value since ancient times (1). Naturally thriving in diverse habitats from rocky slopes to dry grasslands demonstrates remarkable resilience to various soil types and climate conditions. Rich in phenolic compounds and flavonoids, this wild species has also been traditionally valued for its medicinal properties. Its aerial parts have long been used to support digestive and liver health (2). This study analyzed the aqueous leaf extract of wild artichokes collected on the south coast of Sicily (Syracuse, Italy) to characterize their phenolic composition and evaluate their antioxidant capacity through cell-free assays. Subsequently, the crude extract was investigated for its potential chemotherapeutic effects using Caco-2 cells as an *in vitro* model of colorectal carcinoma (CRC).

Folin-Ciocalteu and aluminium chloride spectrophotometric methods were applied to quantify, respectively, total phenolic (TPC) and flavonoid (TFC) compounds, finding values of 55.33±6.55 mg of GAE/g of extract (TPC) and 28.18±0.63 mg of CE/g of extract. Then, the qualitative composition of the extract was further investigated by high-performance liquid chromatography with diode-array detection (HPLC-DAD), revealing the presence of several phenolic acids, primarily caffeoylquinic acid derivatives, along with flavonoids such as luteolin and apigenin derivatives. The extract also demonstrated notable antioxidant activity, as evidenced by the DPPH radical scavenging assay (IC₅₀ = 164.38 ± 2.74 µg/mL) and the catalase-like activity assay (IC₅₀ = 314.73 ± 2.26 µg/mL). In Caco-2 cells, the wild artichoke leaf extract induced a dose-dependent reduction in cell viability (IC₅₀ 24h: 13.07±0.64 µg/mL), associated with modulation of key molecular targets involved in oxidative stress response and apoptosis, including upregulation of HO-1, Bax and p53 protein levels. On the other hand, the extract's mechanism of action did not involve dysregulation of cellular redox homeostasis or mitochondrial functionality, as we did not observe any statistically significant changes in ROS and GSH levels, as well as in mitochondrial activity measured by Seahorse technology. These results contribute to the growing body of evidence supporting the health-promoting properties of this *Cynara* taxon. The complete phytocomplex contained in the leaves appears to offer chemotherapeutic potential against colorectal cancer.



Fig. 1. *C. sylvestris* (Asteraceae) at the collection site. Syracuse, Italy

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7.5 = Saffron flowers: preliminary investigations of a by-product with neuroprotective potential

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During the production process of saffron, a spice consisting only of the stigmas of *Crocus sativus* L., huge quantities of the remaining floral parts, namely tepals and stamens, are discarded. Recently, given the growing interest in phytotherapy for the valorization of by-products of plant origin, saffron flowers have been studied and re-evaluated, as a valid source of active compounds potentially useful in the nutraceutical field.

In this work, the phytocomplex of a sample of organic Tuscan saffron flowers was studied, first through phytochemical investigations aimed at quantifying the total content of phenols (including flavonoids, anthocyanins and flavan-3-ols) and polysaccharides. Furthermore, the anti-radical power was evaluated by means of DPPH and FRAP assays and the bioaccessibility of the active components through a simulated digestion process *in vitro*. In order to analyze the potential neuroprotective action of saffron flower extract (SFE), the SH-SY5Y cell line, a human neuronal model, was used through a series of neuronal stress models (oxidative stress, cortisol stress, serum deprivation). Cell viability and the production of reactive oxygen species (ROS) were then studied.

Although the results obtained are still preliminary and need to be further investigated, we can already state that the saffron flower extract SFE has been shown to possess an interesting phytocomplex, although derived from a by-product, characterized by a fair amount of phenolic and polysaccharide compounds. The high stability of the phenolic component resulting from the digestive process makes SFE concretely exploitable for nutraceutical and health purposes. Saffron flower extract exerts a protective action *in vitro* against various types of neuronal stress. Its antioxidant activity is significant and is expressed both in cell-free assays and *in vitro* on a human neuronal model, lowering the production of ROS.

Through further investigations and studies on the molecular mechanisms underlying the action of the extract, SFE will be evaluated for its protective potential against broad-spectrum neuronal stress, in support of energy metabolism and cognitive functioning.

7.6 = The role of microRNAs and lipophilic compounds in rapeseed pollen-pistil communication

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The current global warming induces intense stress on plant species and critically impacts on their reproduction. Morphological, physiological, and molecular alterations of the reproductive organs have been observed to negatively affect crop performance and yield, with serious economic consequences (1).

In Angiosperms, the ability of anthers to produce and release pollen and the signalling pathways they use to communicate with the pistil are negatively influenced by heat stress (HS), leading to a decrease in pollen viability, germinability, and ability to deliver the sperm cells to the ovule, resulting in plant infertility (2). In this context, HEATSTOP (*Understanding the impAcT of heat STress On Pollen-pistil communication in crops*), a PRIN 2022 PNRR project funded by the European Union (NextGeneration EU - Mission 4, Component 2, Investment 1.1) through the Italian Ministry of University and Research (MUR), proposes an original approach to the issue, focusing on the molecular mechanisms underlying pollen-pistil interactions and how they are invalidated by high daily temperatures during pollen development and release. In detail, the novelty of this project is to achieve a better understanding of the plant fertilization event, with a special focus on the content of pollensomes (PS), the extracellular vesicles released during pollen germination, and the putative function of microRNAs (miRNAs) and lipophilic compounds (e.g., fatty acids) produced by pollen or accumulated in PS. The lack of knowledge about the role of miRNAs during pollination is quite surprising, considering that these short non-coding RNAs are known to extensively modulate gene expression, for example by controlling flowering and stress response (3).

HS-sensitive and HS-tolerant cultivars of rapeseed (*Brassica napus* L.), selected because of the importance of this crop in the global economy, were exposed to high temperature regimes at crucial steps of microsporogenesis for pollen collection. Thus, pollen lipophilic fraction and miRNAs were isolated before and after germination, as well as those from PS, and subjected to Gas Chromatography-Mass Spectrometry analysis and Next Generation Sequencing, respectively, to identify these components and to understand if they could be involved in the regulation of signal transduction pathways related to fertilization and pollen development and activity.

The lipophilic fraction of pollen secretome showed some metabolites that could potentially be involved in the interaction with stigmas, either directly or via PS. In particular, fatty acids seemed to be potential signalling molecules, varying between hydrated and germinated pollen, but also with respect to PS (which could transport them toward the gynoecium), both in stressed and control samples.

Abiotic environmental stressors may influence the synthesis of miRNAs and, consequently, modulate their activity. Thus, the miRNome of pollen and PS, under the various conditions tested, was sequenced and significantly differentially expressed miRNAs were identified. After validation by Real Time-PCR, bioinformatics tools are currently being used to predict the putative mRNA targets of these miRNAs and to carry out a relative Gene Ontology enrichment analysis.

Taken together, these preliminary findings suggest the possibility that both miRNAs and lipids are involved in the molecular mechanisms underlying pollen-stigma interaction, even in the presence of HS, providing clues for the development of biotechnological strategies to improve crop productivity and resilience.

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7.6 = Exploring rhizobacteria-induced molecular pathways in *Solanum lycopersicum*

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Plants coexist and interact with various soil microorganisms, including archaea, bacteria, fungi, and protists, which collectively form the root microbiota. These microbial communities play a pivotal role in promoting plant growth by enhancing nutrient uptake, boosting immunity, and increasing tolerance to environmental stresses. Due to their beneficial effects, soil microorganisms are increasingly regarded as valuable tools for the development of biofertilizers, sparking growing interest in the molecular mechanisms underlying their interactions with plants, particularly crops (1).

In this study, we investigated the responses of *Solanum lycopersicum* cv. Micro-Tom to colonization by potentially beneficial bacteria. We employed molecular and microscopic techniques to evaluate root phenotypes, the molecular pathways involved, and bacterial localization on and within the roots. The bacterial species studied belong to the genera *Bacillus*, *Kocuria*, *Glutamicibacter*, and *Microbacterium*, and were previously isolated by our research group from a commercial compost derived from food scraps and produced by S.E.S.A. S.p.A. (Este, Italy). Characterization of their plant growth-promoting traits revealed their ability to solubilize phosphorus, produce siderophores, and synthesize auxin. Phenotypic analysis of root morphology following bacterial treatments revealed a distinct response of seedlings to *Bacillus sonorensis* (Fig. 1). RNA sequencing analysis of tomato plants inoculated with individual bacterial species identified specific gene expression profiles associated with key plant-microbe interaction pathways, including flavonoid biosynthesis, ethylene signaling, and immune responses (Fig. 2). The transcriptomic data were complemented by metabolomic profiling of tomato tissues and exudates, which revealed signaling molecules and metabolites crucial for the establishment of the plant-bacteria interaction and plant developmental pathways. Finally, bacteria expressing fluorescently-tagged proteins were tracked in tomato organs and tissues using confocal microscopy to characterize their spatial interactions with the plant and to determine whether the bacteria exhibit an endophytic lifestyle within the root (Fig. 3).

This study advances our understanding of root-microbiome interactions and their importance in promoting One Health (2), particularly by contributing to the development of sustainable agricultural practices aligned with the European Union's Sustainable Development Goals for 2030 (3).

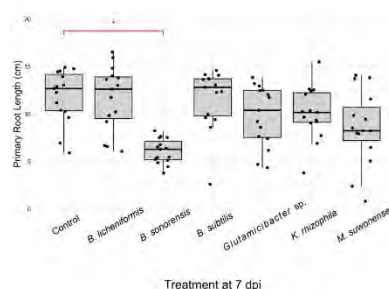


Fig. 1. Length of tomato roots upon single bacteria treatments. Dpi, days post inoculation

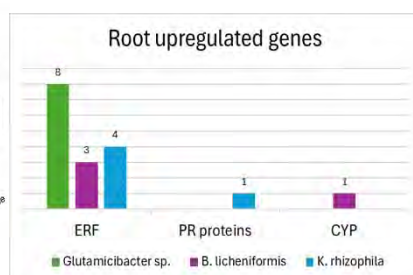


Fig. 2. Upregulated genes in roots upon bacterial inocula. ERF: Ethylene Response Transcription Factors; PR: Pathogenesis-Related proteins; CYP: Cytochrome P450 enzymes

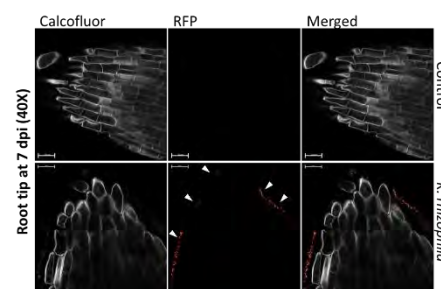


Fig. 3. Localization of *K. rhizophila* expressing RFP (indicated by white arrows) on tomato root at 7 dpi at the confocal microscope. Scale bar, 20 μm

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7.7 = Application of natural deep eutectic solvents for extraction of bioactive compounds from red apple pomace: optimization, phytochemical characterization and biological assays

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The plant-based by-products produced by the food industry have an extensive range of potential for use in several contexts, including functional foods, supplements, and cosmetics, among others. Because of their delicious flavour and recognized health benefits, apples are the only fruit that is consumed in large quantities worldwide. The leftovers of apple processing are collected as food waste, the so-called apple pomace (AP), which is a potentially valuable waste product, as the skin of apples contains a higher concentration of phenolic compounds than the pulp (1), and, in case of red varieties, of anthocyanins.

An attractive approach for the valorisation of apple pomace may be the extraction of bioactive metabolites utilizing innovative technologies. The extraction of bioactive components from apple pomace using NADES (Natural Deep Eutectic Solvents) and MAE (Microwave-Assisted Extraction) was investigated in this study. To assess extraction efficiency in terms of TAC (Total Anthocyanin Content), and antioxidant potential, six NADES mixtures with varying molar ratios were investigated as green extraction solvents. Choline chloride: citric acid (2:1) with 50% water outperformed the other solvents. This NADES mixture underwent additional evaluation in order to optimize the MAE processing parameters through a Response Surface Methodology approach. The solid to liquid ratio (0.01-0.05 g L⁻¹), temperature (30-60°C), and extraction time (5-30 min) were used as X variables in the model, while TAC and the Oxygen Radical Antioxidant Capacity (ORAC) as the Y responses. Among the tested conditions, the highest antioxidant potential value was obtained using sonication time 27.5 min, solid/liquid ratio 0.05, and temperature 51 °C. Liquid chromatography-mass spectrometry (HPLC-MS) and high-performance liquid chromatography with diode array detection (HPLC-DAD) were used to identify and quantify anthocyanins and the other classes of flavonoids for further characterization of optimized extract.

To assess cell tolerance to the selected NADES mixture, preliminary biological assays were conducted using the MTT assay across various cell lines. The determined EC₅₀ values ranged from 0.44±0.1% to 0.61±0.03% for the cancer cell lines MDA-MB-231, MCF-7, SH-SY5Y, and Caco-2, as well as for the non-cancerous cell line MCF10A.

Optimized and non-optimized NADES extracts of AP were tested at different concentrations (from 0.2% to 0.01% v/v) showing no cytotoxicity on all cell lines considered. Anti-inflammatory activity was also studied by the albumin denaturation inhibition test. Using the Bovine Serum Albumin (BSA) assay, the NADES mixture demonstrated intrinsic inhibitory activity on protein denaturation at cell-tolerant concentrations (0.2% to 0.5%). To further investigate anti-inflammatory effects in cellular models of the bioactive molecules, removal of the NADES solvent will be required.

Regarding the antioxidant activity, ROS reduction and intracellular GSH restoration in H₂O₂-induced oxidative stress cells will be evaluated. Results will contribute to provide a preliminary picture of the potential application of red apple pomace.

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7.7 = Beyond tradition: A phytochemical and bioactivity study of "Genepi" (*Artemisia* spp.) preparations

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Artemisia genipi Weber and *A. umbelliformis* Lam., collectively known as “Genepi,” are alpine species traditionally used to prepare herbal beverages with reputed health benefits (1). While the composition of their essential oils is well studied, comprehensive chemical profiling of hydroalcoholic extracts is limited (2), and the composition of aqueous infusions remains largely unexplored. This study investigated both volatile and non-volatile components of “Genepi” infusions and hydroalcoholic extracts using SPME-GC/MS and HPLC-DAD/LC-MS techniques. Monoterpenoids, particularly α -thujone and β -thujone, dominated the volatile fraction of infusions, whereas sesquiterpenoids such as reynosin and ambrosin were more prevalent in hydroalcoholic extracts. Among phenolic compounds, tricin derivatives, luteolin, and axillarin were most abundant.

Beyond phytochemical characterization, selected aspects of bioactivity were evaluated:

- (i) antiradical activity using ABTS•+ and DPPH• assays;
- (ii) effects on cell viability in wild-type (HWT) muscle cell lines.

Infusions exhibited stronger radical scavenging activity than hydroalcoholic extracts in both ABTS•+ and DPPH• assays. In vitro, hydroalcoholic extracts of *A. genipi* demonstrated dose-dependent cytotoxicity, whereas infusions were non-toxic and enhanced cell viability, with *A. umbelliformis* infusion (0.05 mg/mL) increasing viability by up to 30%. Under myogenic differentiation conditions, both extracts supported myotube viability at low concentrations. Furthermore, *A. umbelliformis* infusion significantly downregulated IL6 expression, suggesting potential anti-inflammatory and immunomodulatory effects.

These findings offer novel insights into the bioactive profiles and biological activities of “Genepi” preparations, supporting their traditional medicinal use and informing future therapeutic applications.

Additional anti-inflammatory and antioxidant assays are ongoing to further elucidate the health-promoting potential of these plant extracts.

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7.7 = ¹H NMR-based metabolomics provides new insights into chemodiversity, biochemistry, and response to abiotic stimuli of *Cladonia* lichens from dry grasslands

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Lichens are complex organisms, defined as a self-sustaining ecosystem generated from the symbiosis between a mycobiont and a photobiont (1). The interest in these fascinating organisms lies both in their bioactivities, potentially useful for human beings, and in their multiple roles in ecosystems (2). They are characterized by structures and metabolites not found in the interacting partners if cultivated separately, and the understanding of molecular and physiological processes occurring within the symbiosis is still an ongoing challenge.

This study investigates four species of the genus *Cladonia* (*C. foliacea*, *C. polycarpoides*, *C. rangiformis*, *C. rei*), endowed with numerous bioactivities (2) through ¹H NMR metabolomics. Each species was sampled once a month for one year, in 5 different sites in dry grasslands of the Ticino River valley (Italy). The objectives were: i) to acquire and elucidate for the first time the ¹H NMR profile of these species, ii) to picture intraspecies metabolomic differences, and iii) interspecies metabolomic variation due to environmental factors.

Using chemometrics, several validated models were obtained. First, these models provided a tool to discriminate between the four examined *Cladonia* species. Then, they revealed how these species respond to environmental stimuli. The PLS-DA model (Discriminant Class=Species) showed that species clustered not only based on their biomarkers (i.e., usnic, protocetatic, fumarprotocetatic, rangiformic, norrangiformic, sekikaic, and homosekikaic acids), but also for the enrichment in primary metabolites. For instance, fumarate and sucrose were more concentrated in *C. polycarpoides*. *C. rangiformis* had the highest concentration of arabinitol, acetate, and alanine, while glucose and trehalose were more abundant in *C. rei*.

Several intraspecies differences, both related to time-point and harvesting site, were also detected. In particular, the OPLS model (y variable=temperature) revealed that the metabolome of all species was significantly affected by the temperature gradient. Arabinitol emerged as one of the most important metabolites produced in response to chilling; on the contrary, increasing temperature resulted in the accumulation of a sugar alcohol identified (by 2D NMR and ToF-MS) for the first time in lichens, the β -glucopyranose(1→2)mannitol. Moreover, the same OPLS model, provided information on the nitrogen cycle in *Cladonia*, letting hypothesize that nitrogen is assimilated into glutamine during winter and released in spring-summer, favoring lichen catabolism and reproductive activity. The cycle potentially involves threonine, β -glucopyranose(1→2)mannitol, and arabinitol as further key metabolites. What was observed suggests a highly integrated nitrogen-carbon regulatory mechanism tuned to environmental changes.

The overall results offer a metabolomics-based discrimination method for *Cladonia* species, bringing insights both on when to harvest these lichens in view of a specific medicinal use and on how they might adapt to environmental changes.

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7.7 = Phytochemical and Biological Characterization of *Pinus pinea* Bark: a Widely Available but Overlooked Source of Antioxidant Compounds

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Pine bark is well known for its abundance of bioactive compounds and promising therapeutic properties, particularly due to its antioxidant activity. Considerable research has been devoted to species such as *Pinus pinaster*¹ and *Pinus gerardiana*², which are rich in polyphenols and exhibit strong free radical scavenging capacity. In contrast, the bark of *Pinus pinea* L. (stone pine) remains largely underexplored. Although this Mediterranean species is economically important—especially for its edible seeds—and has a long history of use in traditional medicine, scientific studies on the phytochemical composition and antioxidant potential of its bark are notably scarce.

This study examined the bark of *Pinus pinea* L. collected from a mountainous area near Brescia (Italy). A hydroalcoholic solvent was used for extraction, and the resulting extract was lyophilized for further analysis. Phytochemical profiling was conducted using HPLC-DAD and UHPLC-QToF, while the inorganic composition was assessed using TXRF. The biological activity of the extract was assessed in the SH-SY5Y human neuroblastoma cell line by performing cytotoxicity assays and evaluating its protective effect against induced oxidative stress.

The phytochemical analysis revealed a high concentration of phenolic compounds, with taxifolin representing approximately 20% of the total extract. Taxifolin is a well-characterized flavonoid with documented anticancer, antioxidant, and anti-inflammatory properties. TXRF analysis showed the presence of several trace elements, including nickel, zinc, copper, and iron.

Biological assays demonstrated a protective effect of the extract against oxidative stress in SH-SY5Y cells, likely due to its rich phenolic content.

In conclusion, although stone pine is a widely distributed species in Italy and throughout the Mediterranean, its bark has received little scientific attention. This study reveals its considerable phytochemical richness and noteworthy biological activity, emphasizing the need for further research on this underutilized yet readily available natural resource.

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7.7 = Pre-Harvest Heat Stress Modulates Post-Harvest Functional Traits, Metabolic Profiles, and Gene Expression During Cold Storage of *Diplotaxis tenuifolia*

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Wild rocket salad (*Diplotaxis tenuifolia*, L.) is an herbaceous plant belonging to the Brassicaceae family valued for its pungent flavour, a distinguishable aroma and nutritional properties, yet its post-harvest storage remains limited (1). To extend shelf life, leaves are typically stored under refrigeration, which introduces a combination of abiotic stresses such as low temperature, darkness, and dehydration. Concurrently, rising global temperatures and the increasing frequency of heatwaves are imposing additional stress during cultivation. This study investigates whether thermal stress experienced prior to harvest influences the physiological and molecular responses of rocket leaves during cold storage.

Plants were subjected to a short-term heat treatment (35 °C for 3 days) at the pre-harvest stage, while control plants remained at 22 °C. Harvested leaves were then stored at 6 °C for 7 days. We assessed the impact of pre-harvest heat exposure on post-harvest leaf damage, volatile organic compound (VOC) emissions, metabolite composition, and transcriptomic profiles. Leaves from heat-stressed plants exhibited greater cellular damage, evidenced by reduced chlorophyll content and elevated ion leakage. VOC and metabolome profiles revealed persistent alterations in aroma and biochemical profiles after chilled storage. Transcriptome analysis identified differential expression of over 900 genes after pre-harvest heat treatment and storage under cold conditions. Gene Ontology (GO) term and Kyoto Encyclopedia of Genes and Genomes (KEGG) analyses highlighted enrichment in pathways related to stress signaling, secondary metabolism, and hormone responses. Furthermore, parallels with known dark-storage responses in *Arabidopsis thaliana* were noted, including activation of senescence-associated processes such as chlorophyll degradation and macromolecular recycling (2). Integration of VOC and gene expression data suggests the involvement of post-transcriptional regulatory mechanisms (3).

These findings underscore the lasting influence of pre-harvest environmental conditions on post-harvest quality and stress resilience, offering insights for optimizing crop management and storage strategies under climate variability.

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7.8 = Nanoencapsulated oils from plants of ethnobotanical interest as a sustainable approach to modify the lipid profile of *Tenebrio molitor* (Tenebrionidea)

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Animal-derived foods represent a crucial source of essential amino acids. Considering the rapid population growth projected in the next years, meeting the demand for proteins could become unfeasible from both economic and ecological points of view. To address this rising demand, it is necessary to enhance the sustainability of animal production through the adoption of innovative livestock feeding strategies. Among novel feed ingredients, insects represent a valid alternative choice due to their high nutritional value and ease of rearing. In particular, *Tenebrio molitor* larvae (or yellow mealworm larvae, YML) have been recognized by the European Union as insects with the highest potential for both food and feed resources¹.

In this work, the modification and improvement of YML lipid profile using nanosystems produced from plant-derived by-products were investigated. Exopolysaccharides produced by the cyanobacteria *Cyanospira capsulata*, or phenols present in olive pomace, were added to a natural lipid mixture represented by egg-yolk phosphatidylcholine. The combination of these elements allowed the modification of the bending rigidity of the polysaccharides, creating smaller nanoparticles capable of encapsulating oils extracted from *Sclerocarya birrea* and *Perilla frutescens*. These two plants show ethnobotanical value, and both contain lipids of interest in enhancing the nutritional profile of YML.

The physicochemical properties of the produced nanoformulations were tested to obtain information about their dimensions, surface charge, and structure. Nanosystems loaded with *P. frutescens* oil showed an average size of 150-200 nm, while nanoformulations with *S. birrea* oil were in the range 200-300 nm. In both cases, the surface charge was around -70 mV, remarkably lower than their plain counterparts (~ -25 mV). From *Small Angle Scattering* measurements the nanovectors showed a globular shape characterized by an oil-rich core.

The prepared nanoformulations were administered through the diet to *T. molitor* larvae for three weeks, after which *Gas Chromatography Mass Spectroscopy* (GC-MS) was performed on the obtained YML flour and nanosystems to investigate the lipid composition of the vectors and the variations induced in the lipid profile of the insects.

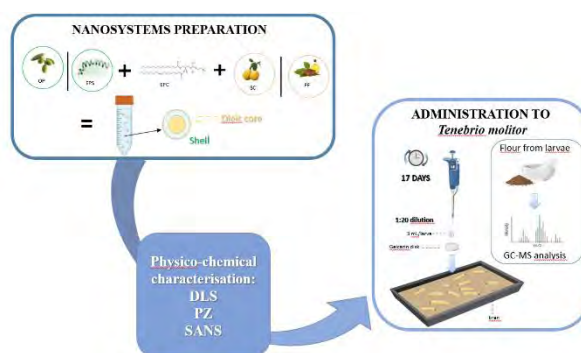


Fig. 4. Experimental design

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7.8 = Heat stress responses in four freshwater microalgal species: *Chlorella vulgaris*, *Desmodesmus abundans*, *Nostoc sp.*, and *Anabaena sp.*

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The continuous production of greenhouse gases, particularly CO₂ generated by the combustion of fossil fuels, has had a significant impact on the environment, often generalized under the term "global warming". Aquatic ecosystems, like terrestrial ones, are susceptible to climate change. Global temperatures are predicted to increase between 1.4°C and 5.8°C over the next 100 years. However, in high-latitude regions, the temperature rise could reach up to 10°C (1). Among various aquatic ecosystems, lakes—especially shallow ones—are most affected by climate change due to their high surface-to-volume ratio, making them more hydrologically sensitive (2). This study focuses on variations in morphological and biochemical characteristics associated with heat stress tolerance in four freshwater microalgal species: two green microalgae, *Desmodesmus abundans* and *Chlorella vulgaris*, and two cyanobacteria, *Nostoc sp.* and *Anabaena sp.* The aim is to safeguard ecosystems and conserve biodiversity, with potential biotechnological applications. Cultures were divided into three groups: control cultures maintained at 23°C, and stressed cultures grown at 28°C and 33°C. All cultures were exposed to 2000 lux light intensity and a 16-hour light/8-hour dark photoperiod. Cell growth was analyzed by counting cells in a Bürker chamber under an optical microscope. Both control and stressed cultures were observed under a confocal microscope using colorants such as Fluorescein Diacetate (cell viability marker) and Nile Red (neutral lipid marker). After freeze-drying, cultures underwent biochemical analyses to analyze any changes in both primary and secondary metabolites. Following heat stress, a reduction in cell density was observed in all microalgae, particularly at 33°C, with *Chlorella vulgaris* showing the lowest reduction. Morphological analyses revealed an increase in cell diameter in *Desmodesmus abundans* and *Nostoc sp.* under heat stress, and a decrease in the number of cells per filament in both cyanobacterial species. Biochemical analyses of chlorophylls and carotenoids showed species-specific differences, indicating different responses to heat stress. Preliminary results suggest that *Chlorella vulgaris* is the most heat-tolerant species among those analyzed. This study demonstrates that different microalgal species respond uniquely to heat stress, implying that climate change will affect species in diverse ways over time.

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7.8 = Reproductive challenges and allergenic shifts in *Malus domestica* facing temperature stress

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This study analysed the impact of temperature stress on the reproductive biology and allergenic protein expression of *Malus domestica*. *M. domestica* cv. Florina plants were subjected to temperature stress in order to simulate the impact of a late spring frost during flowering — a phenomenon that is a common cause of crop loss. The experimental group was exposed to a temperature of 1 °C for 12 hours after anthesis, while the control group was kept at 20 °C. All plants were manually self-pollinated prior to treatment. Due to the species' gametophytic self-incompatibility, the experiment aimed to evaluate potential pollen-pistil interactions and incompatibility mechanisms under thermal stress. The pistils were collected 48 hours after pollination, immediately frozen in liquid nitrogen and stored at -80 °C. Some of the pistils were fixed in 70% ethanol for immunohistochemical analysis and evaluation of pollen tube growth.

The effects of temperature stress on pollen were studied *in vitro*: under thermal stress conditions, the cytoskeletal organisation of pollen tubes is altered. Pollen grains (PGs), extracellular vesicles (EVs) and a vesicle-free medium (EVF) were separated by filtration and differential ultracentrifugation (1). The pollen granules in EVs were quantified using nanoparticle tracking analysis and stained with FM 4-64 to confirm their integrity. Total proteins were then isolated from the three fractions, quantified and resolved by SDS-PAGE. Immunoblotting was then used to verify the presence of the following: (I) pollen markers (e.g. ALIX homologues and clathrin), confirming their presence in EVs; (II) markers of other cellular compartments; and (III) profilin and actin. Finally, the presence of actin-bound profilin in EVs or EVFs was assessed by co-immunoprecipitation.

The secretion of profilin within pollensomes suggests a role in distant signalling, which could lead to cytoskeletal remodelling of pistil cells. The secretion of pollensomes and profilin increased in response to temperature stress.

Finally, Mal d 1, the main allergen in apples, was quantified in the epicarp and mesocarp of *M. domestica* fruits stored at 1 °C. The fruits were harvested on four dates (17, 24 and 31 October and 6 November), and for each date, the samples were analysed in triplicate, with each replicate consisting of three individual fruits. These data will help us to understand how harvest times and storage conditions influence the accumulation of allergens in apple tissues.

Together, the results shed light on the multifaceted effects of temperature stress on *Malus domestica*, revealing its influence on pollen-pistil interactions, cytoskeletal dynamics, and allergen expression—factors that are crucial for both crop yield and consumer health.

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7.8 = TOR inhibitor and autophagy inducer Rapamycin as a promoter for salt stress resistance in *Eruca sativa* seedlings

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Salt stress is one of the most common factors reducing the productivity of crops. We tested the effect of Rapamycin, an mTOR inhibitor and autophagy inducer, on salt-stressed *Eruca sativa* at the seedling stage. We analyzed the germination rate, the macromorphology of seedlings, the micromorphology, and the ultrastructure of the cotyledons with light microscope and Transmission Electron Microscope (TEM), respectively. In Rapamycin-treated samples, results revealed a reduced impact of salt stress on germination and plant development in terms of dry weight production and stem elongation. Moreover, Rapamycin on salt-stressed plants, hits positively anatomical features such as intercellular space and leaf thickness. Lastly, plants treated with rapamycin showed a reduction in the percentage of lipid droplets. TEM observation highlighted induced autophagic activity in Rapamycin-exposed samples, along with increased membrane recycling. We also observed a negative effect in germinating seeds and seedlings in *E. sativa* treated with DMSO, the solvent normally provided with Rapamycin. Therefore, DMSO as a solvent does not appear to have a neutral effect on *E. sativa* plants. Salt stress (with DMSO) appears to reduce the mobilization of the lipid droplets stored in the embryo of *E. sativa* which, consequently, remains in the developing seedlings until a late stage. Based on the above-cited results, Rapamycin showed a protective effect on seed germination and development in salt-treated *E. sativa* samples, especially by ensuring the lipid storage mobilization.



Fig.1. Morphological aspect of *E. sativa* plants for each treatment

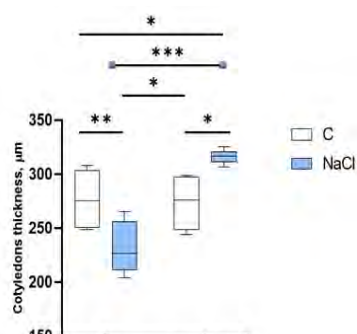


Fig. 2. Analysis on cotyledon thickness



Fig. 3. TEM micrograph of *E. sativa* cotyledon

7.8 = Isotopic insights into heat stress resilience: comparing chlorophyll-deficient wheat lines during the 2022 heat wave

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After a cold start to spring, in 2022 an anomalous heat wave occurred in May in Northern Italy well above the range of climatological normality. A hypothesis in the scientific literature states that pale green crops may exhibit higher heat resistance than the corresponding wild types because the light reflection by pale green leaves might also mitigate the effects of heat waves (1). The availability of eight near-isogenic durum (LD222 and its mutants ANDW-7A, ANDW-7B, ANDW-8A) and bread wheat lines (Novosibirskaya-67 and its mutants ANBW-4A, ANBW-4B, ANK-32A), grown under controlled watering conditions at the Botanical Garden of Ferrara (2), provided an opportunity to investigate the response of wheat lines with different chlorophyll accumulation capacities to high temperatures without the interference of drought stress. Carbon isotopic analysis, often coupled with nitrogen isotopic analysis, is an elective, extremely sensitive method to trace plant susceptibility to environmental stress (3). Therefore, in this study we compared the wheat lines, focusing on carbon and nitrogen contents and isotopic ratios ($\delta^{13}\text{C}$ and $\delta^{15}\text{N}$) in leaves and grains, along with grain productivity.

Results showed that while carbon and nitrogen concentrations remained stable across genotypes, $\delta^{13}\text{C}$ values were consistently lower in leaves of chlorophyll-deficient mutants compared to their wild-type counterparts, indicating increased carbon isotope discrimination. However, this trend did not persist in grains, except for the ANBW-4A bread wheat mutant. The decrease in $\delta^{15}\text{N}$ caused by metabolic fractionation was evaluated from the soil value (5‰) to leaves and grains. $\delta^{15}\text{N}$ values decreased significantly only in the leaves of durum wheat mutants compared to wild-type LD222. Correlation analyses revealed that, while isotopic ratios were linked to the leaf chlorophyll content, they were not predictive of grain productivity. Nonetheless, the best-performing genotypes in terms of grain filling, wild-type durum wheat LD222 and bread wheat mutant ANBW-4B, exhibited similar $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ values.

We conclude that the hypothesis of superior heat stress resistance in pale green wheat lines is overly simplistic and should not be blindly accepted. Among the examined lines, ANBW-4B deserves further investigation to assess its potential superior resistance to environmental stresses in the context of climatic changes.

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7.8 = Effects of different abiotic stresses on *Phaeodactylum tricornutum*: perspectives for the large-scale production

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Microalgae produce high-value molecules, such as proteins, lipids and polyphenols, and can be exploited for multiple biotechnological applications, such as feed for aquaculture. Diatoms, among microalgae, are known for the production of a porous silica cell wall, but they can also produce biomass rich in fatty acids, including essential ones, and fucoxanthin, a carotenoid with known antioxidant activity. Metabolite production and accumulation can be affected by environmental conditions, making it possible to design culture conditions to increase the production of specific metabolites or biomass. For example, changes in light quality and quantity have been reported to affect both pigment profile and growth, with blue light increasing lipid production in some microalgae, while red light has a positive effect on biomass production in other species [1]. In our work, we investigated the impact of different abiotic stresses on *Phaeodactylum tricornutum*, a peculiar diatom that exhibits the ability to change morphotype under different environmental conditions, with only one morphotype being silicified [2]. The scale-up of the cultures was also tested as a preliminary step toward actual industrial applications.

P. tricornutum CCAP1055/4 was cultured under three different light setups (White, Red-enriched and Blue-enriched LED light) in 300 mL of undiluted or 10% diluted artificial seawater enriched with f/2 medium. Sodium metasilicate pentahydrate was added at a final concentration of 100 µM as a source of silica. Growth was monitored through cell count. Total polyphenols, total proteins, pigments and fatty acids profile were evaluated to assess biomass composition after 14 days of cultivation. Moreover, morphological changes were observed using transmission electron microscopy (TEM). Preliminary tests on the scale-up of the cultures were carried out to test different designs of photobioreactors, including a cylindrical photobioreactor and an open-pond-like setup.

Our results showed that growth was negatively affected by reduced salinity, while it was enhanced by red light. Interestingly, cultures grown under red light in 10% diluted artificial seawater reached the same concentration as the ones grown in undiluted medium with red light, meaning that the positive effect of these wavelengths on growth completely balanced the effect of reduced salinity. However, cultures grown with red light, regardless of salinity, showed the lowest concentrations of both proteins and polyphenols per cell among all cultures. Blue light, conversely, had no impact on the total protein content and biomass production, but led to an increase in saturated fatty acids in the cultures grown in undiluted artificial seawater. TEM images revealed a major presence of lipid droplets in cells cultivated in reduced salinity with white light, but no significant morphotype change was recorded in any of the tested conditions. An increased duration of the initial lag phase was recorded when culturing in larger volumes, but, generally, cultures still reached a plateau after 14 days. These tests serve as the initial step in selecting the optimal environmental conditions for various biotechnological applications, such as the large-scale production of *P. tricornutum* as a feed for aquaculture.

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7.8 = Evaluation of epigenome and rhizosphere microbiome changes in *Solanum lycopersicum* L. treated with Gadolinium

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Gadolinium (Gd) is a lanthanide belonging to the Rare Earth Elements (REEs) group, which are widely used in various anthropogenic applications (*e.g.* automotive industry, glass and ceramic production, military equipment, medicine, agriculture, etc.). Mining activities, improper disposal and environmental dispersion have led to a notable increase in REEs concentration. In particular, Gd is employed in medicine as a contrast agent in magnetic resonance imaging, leading to surface water contamination. The effects of REEs on living organisms and on the environment remain unclear, therefore many studies are focusing on the morphological, physiological and genetic changes they may induce. This study aimed to assess the effects of Gd on *Solanum lycopersicum* L. (cultivar MicroTom) grown in a hydroponic system. After germination, tomato plants were transferred to two different hydroponic systems: one with standard Hoagland solution (control plants), and the other one containing a modified Hoagland solution with Gd 150 μ M. Plants were collected from each hydroponic system (control and Gd 150 μ M.) after one month of treatment (sampling I) and at the end of the life cycle (approximately three months; sampling II). In order to gain a deeper insight into the molecular and epigenetic basis underlying the response of tomato plants to Gd treatment, leaves and roots were analyzed through Methylation Sensitive Amplification Polymorphism (MSAP) method coupled with Next Generation Sequencing (NGS). At the same time, bacterial and fungal communities of the rhizosphere of plants (control and Gd 150 μ M) were characterized through NGS technique. The MSAP results showed that the methylation status was affected by the exposure to Gd. The MSAP-NGS approach allowed us to identify genes that were differentially methylated among the different tomato plants and the two samplings and link them to specific biochemical pathways. Instead, microbiome analysis revealed that the bacterial and fungal community compositions were affected by Gd exposure, resulting in a statistically significant increase of the biodiversity respect with the control. These findings suggest that MicroTom is a good model plant to study the effects of Gd and identify epigenetic variations related to stress, defense mechanisms and tolerance.

7.8 = Response of different rice (*Oryza sativa* L.) genotypes to inorganic arsenic exposure: Carnaroli vs Zhonghua 11

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Rice (*Oryza sativa* L.) is a staple food and one of the most important grain crops worldwide. Italy is the leading rice producer in Europe, accounting for more than fifty percent of the total production and more than two-hundred varieties. Italian rice production is gathered in the regions of Piedmont and Lombardy. In particular, the province of Vercelli is the largest producer of the Carnaroli variety (*O. sativa* subsp. japonica) which originated in Lombardy eighty years ago from a breeding between the Vialone and Lencino varieties. The Carnaroli cultivar is still the most cultivated today and is exported all over the world thanks to nutritional and organoleptic properties of its grains. However, this variety has not yet been sufficiently studied in terms of tolerance to arsenic (As) of the plant.

Rice is one of the main As exposure food sources for humans. Inorganic arsenic (iAs) is steadily present in paddy fields wherein it is easily absorbed by plant roots and, gathering in these organs, alters their development as well as hormonal homeostasis ⁽¹⁾. Paddy rice is more efficient in iAs uptake than other cereals and, although most of this toxic element is sequestered in the root, it also moves up the plant to the edible fruits. The content of iAs in Italian rice varies greatly due to complex interactions between physico-chemical soil properties, agronomic practices, and rice cultivars. ⁽²⁾ Given its toxicity, authoritative organizations such as the European Commission have established strict guidelines and new limits (EU 2023/465) to reduce the iAs presence in rice-based foods, as a measure to counteract its harmful effects in humans. Nowadays, it would be essential to select rice varieties capable of excluding or limiting As accumulation in the grains.

The aim of this research is to deepen the knowledge on the tolerance to iAs of different rice genotypes, particularly Carnaroli variety. Seeds of Carnaroli were sown *in vitro* in presence, or not, of arsenite (As^{III}) or arsenate (As^V), added at sublethal concentrations to the culture media. After ten days, rice seedlings were analyzed at the morphological, cytohistological and chemical levels. The responses of this variety were compared with those of the As-sensitive Zhonghua 11 (*O. sativa* subsp. japonica) ⁽³⁾.

The results indicate that the Carnaroli seedlings showed a greater tolerance to iAs, despite their higher root uptake compared to Zhonghua 11. Lateral root (LR) production was significantly reduced in Carnaroli only when treated with As^{III} compared to the Control, while this reduction was significantly higher in Zhonghua 11 with both iAs forms. Histological analysis of adventitious roots, the main components of the *O. sativa* root system, showed abnormalities in LR development, confirming the results of morphological analysis. Further analyses are currently underway to understand in depth the mechanisms that allow the different rice genotypes to cope with the presence of iAs in the substrate and to evaluate the involvement of the shoot in the sequestration and detoxification of this dangerous metalloid.

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7.8 = Investigating quinoa's plant growth-promoting bacteria and their ability to promote salinity tolerance in *Arabidopsis thaliana*

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Beneficial plant microbes, especially Plant Growth-Promoting Bacteria (PGPB), are essential for host growth and resilience to biotic and abiotic stressors. Seed endophytic bacteria, less studied than rhizobacteria, may significantly enhance germination, seedling establishment, and stress tolerance. Quinoa (*Chenopodium quinoa* Willd.), a native crop from the Andes that is rapidly expanding worldwide, exhibits remarkable tolerance to high salinity **(1)** making it an excellent candidate for investigating beneficial microbes associated with it. Based on the premise that PGPB contribute to host stress resilience **(2)**, this study aimed to isolate and characterize endophytic bacteria from quinoa seeds and roots, and from the seed processing residue (quinoa hull powder, QHP), evaluate their plant growth-promoting (PGP) abilities, and investigate the effect of a selected isolate on the salt-sensitive model plant *Arabidopsis thaliana*. Bacterial isolates from various quinoa matrices were screened for multiple PGP traits, including P and K solubilization, siderophore production, ACC deaminase activity, and NaCl tolerance. Identification was performed by MALDI-TOF. Strain A7A, a *Pseudomonas* sp. isolated from whole seeds of quinoa var. 'Pandela', exhibited the highest PGP activity score and was, therefore, selected for co-culture experiments with *A. thaliana* Col-0. Seedlings were grown with or without 100 mM NaCl and inoculated with isolate A7A. Morphological (fresh weight, root architecture), biochemical (proline, oxidative stress), and transcriptional analyses were conducted. Salinity significantly reduced *Arabidopsis* fresh weight and primary root length. While A7A inhibited growth on non-saline medium, it restored seedling fresh weight to control levels (no salt, no bacteria) under salt stress. A7A also enhanced lateral root number in non-saline conditions and restored it under salt stress. Proline content was dramatically increased by A7A, both in the presence and absence of NaCl. Under salt stress, expression of *AtANR1*, the gene encoding the transcription factor ARABIDOPSIS NITRATE REGULATED1 involved in seed germination and the salt tolerance-related gene *AtSOS1* were upregulated by A7A. These results suggest that endophytic bacteria from stress-tolerant quinoa, like *Pseudomonas* sp. A7A, have the potential to improve salt tolerance in sensitive plants **(3)**, possibly through mechanisms involving increased proline accumulation, modulation of root architecture, and changes in expression of germination- and stress-related genes.

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7.8 = Mechanosensitivity in Plants: Vibrational Signal Perception and Response in *Arabidopsis thaliana*

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The PRIN DAMATIRA project explores how plants perceive and respond to acoustic and vibrational cues, integrating multidisciplinary approaches and methodologies. We utilized laser Doppler vibrometry to record vibrational signals produced by *Eurydema oleracea* during feeding on *Arabidopsis thaliana* leaves, and to determine an average resonance frequency that maximizes mechanical deformation at the leaf level.

Based on these data, we developed two distinct artificial vibrational stimuli: the first emulates the feeding activity of *E. oleracea* on the abaxial surface of *A. thaliana* leaves, while the second is designed to induce mechanical deformation by exploiting the leaves' resonant properties. Both stimuli were initially characterized through finite element analysis (FEA) to model their biomechanical effects, and subsequently applied in vivo using a custom vibrational platform to evaluate plant responses at biological, biochemical, and molecular levels.

A key technological advancement of the project was the use of X-ray micro-computed tomography (micro-CT) to generate realistic three-dimensional reconstructions of *A. thaliana* under live, non-destructive conditions. This represents a pioneering application of micro-CT to living, non-woody plants, with the optimization of acquisition parameters opening novel perspectives in clinical and structural botany.

To assess the effects of vibrational treatments, we analysed several morphometric traits (germination rate, fresh biomass, leaf number, primary root length), biochemical markers (total phenolic content and antioxidant activity), and gene expression profiles. The molecular analysis focused on genes associated with cell wall remodelling (XTH11, XTH19, XTH29, PME12, PME18, PME34) and mechanosensation (PIEZO1, MSL2, MSL6, MSL9).

In addition, carotenoid content and volatile organic compound (VOC) profiles were examined in plants exposed to insect-derived vibrations and are currently under investigation for artificially stimulated samples as well. This integrated, multi-level approach offers new insights into the physiological and molecular mechanisms underlying plant responses to mechanical stimuli, contributing to the broader understanding of plant mechanosensitivity.

7.8 = *Arabidopsis thaliana* salt stress mitigation by priming with a quinoa seed by-product: linking phytochemical composition to antioxidant defence and gene modulation

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Biostimulants derived from recycled waste or by-products represent a promising eco-friendly technology to help plants cope with abiotic stresses like salinity, which significantly impacts crop production and is predicted to reduce cultivable land globally. Quinoa (*Chenopodium quinoa*), a highly nutritious grain (1), produces large amounts of a by-product rich in saponins (2) and other bioactive compounds during the dehulling process. This study analyzed the potential of this waste as plant biostimulant. The objective was principally to evaluate the stress-protective activity of quinoa hull powder (QHP) on the salt-sensitive model plant *Arabidopsis thaliana*, by assessing its effects on growth, biochemical parameters, and gene expression under control and saline conditions. The phytochemical characterization of QHP revealed that it was rich in saponins, potassium, proline, spermidine (3), carotenoids, and polyphenols, and that it possesses antioxidant capacity. *Arabidopsis* was subjected to salt stress (25, 50, 100 mM NaCl) using two application methods: (i) seed priming with QHP extract prior to sowing, and (ii) transfer of seedlings to QHP-supplemented media. Germination, seedling growth (biomass, root length), oxidative stress markers (MDA, H₂O₂), and transcript levels of stress-responsive genes were measured. The results showed that seed priming with 0.05 mg/mL QHP recovered germination under 50 and 100 mM NaCl. Supplementation of media with 0.05 mg/mL QHP improved shoot and root biomass and primary root length in seedlings under saline conditions. QHP reduced salt-induced oxidative stress to control levels. RT-qPCR analysis revealed that QHP alone induced expression of some stress-responsive genes (e.g., antioxidant enzymes, pathogenesis-related proteins), thereby acting as a priming agent, and modulated the salt-induced expression of others, often attenuating the salt response. In conclusion, the phytochemical analyses showed that QHP is a valuable biowaste rich in stress-protective compounds able to enhance plant tolerance to abiotic stresses. This study also provides, for the first time, insight into the molecular mechanisms underlying QHP activity, such as modulation of the expression of genes involved in antioxidant defense and stress responses. The results support its potential use for crop protection in sustainable agriculture.

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7.8 = Fine soils from dismissed bauxite mines in Puglia (Southern Italy) and selected soil-borne fungi interactions

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The impacts of soil samples containing toxic metals (e.g., As, Ba, Co, Cr, Cs, Cu, Ga, Hf, Mo, Nb, Ni, Rb, Pb, Sr, Th, U, V, Zn, Zr) and Rare Earth Elements (REEs: Ce, Dy, Er, Eu, Gd, Ho, La, Lu, Nd, Pr, Sm, Sc, Tb, Tm, Y, Yb) were evaluated against the phytopathogenic fungi *Fusarium oxysporum* f.sp. *lycopersici* (FOL), *Sclerotinia minor* (SM), *Verticillium dahliae* (VD) and *Trichoderma harzianum* (TH) a biological control agent of plant pathogens. Soil samples were taken from San Giovanni Rotondo, Otranto and Spinazzola dismissed bauxite mines in the Puglia region in the area near (GB2, OD3, SB3) and far (GE2, OE4, SE4) from each mine entrance. Soils were characterized by major and minor elements (1) and REEs (2) composition. Pollutant Total Concentration (PTC), the modified contamination degree index (mCI_d) and the Nemerow Integrated Risk Index (NIRI) were calculated (Table 1) as indicators of risk assessment (3). Soil samples were assayed at 1% in Petri dishes containing Potato Dextrose Agar (PDA) and as liquid cultures in half-strength Potato Dextrose Broth. The percentage of radial growth inhibition, mycelia biomass production and the sporulation inhibition rate were evaluated. The antagonistic activity of the TH mediated by secondary metabolites was also analysed. The Pearson correlation coefficient was estimated among the PTC, mCI_d or NIRI values and each ecological effect on tested fungi.

In PDA experiments, all soil samples lowered VD (21.9-32.2 %), and SM (4.3-65.2 %) while enhancing FOL (73.9-86.6 %) radial growth. The TH mycelial growth was differently altered: reduced by GB2 (25.9 %) or SB3 (22.1 %) and enhanced (9.4-17.1 %) by the other soil samples. Soil samples in PDA altered the sporulation rate of FOL, TH and VD and had no effect on SM microsclerotia differentiation.

In liquid cultures, almost all soil samples affected mycelial biomass production, reduced VD (85-90.2%) and TH (20-70%) conidia production, and increased FOL (35-188%) sporulation. The SM strain did not produce microsclerotia in any liquid media tested. Soil samples in the liquid media also affected TH antagonistic activity mediated by secondary metabolites.

Pearson correlation coefficient correlates the recorded biological effects with PTC, mCI_d and NIRI and indicates a possible dominant role of Rare Earth Elements. These results suggest that the tested soil samples affect the vegetative and reproductive activity and metabolism of the tested soil-borne fungi. The data recommend soil surveys considering the impact on biota, especially in sites of naturalistic, geological, and tourist interest.

Table 1. Pollutant total concentration (PTC), modified contamination degree index (mCI_d) and Nemerow Integrated Risk Index (NIRI) of soil samples collected from dismissed bauxite mines in San Giovanni Rotondo (GB2, GE4), Otranto (OD3, OE4), and Spinazzola (SB3, SE4)

Parameter	Soil samples ^a					
	GB2	GE4	OD3	OE4	SB3	SE4
PTC (g kg ⁻¹ dw)	1.815 ± 0.88	1.207 ± 0.46	3.426 ± 0.19	3.126 ± 0.32	4.376 ± 0.68	3.439 ± 0.98
mCI_d	3.891±0.52 ^d	1.999±0.20 ^e	5.324±0.21 ^b	4.555±0.07 ^c	10.448±0.94 ^a	6.703±0.46 ^b
NIRI	2.934±0.27 ^c	2.934±0.27 ^c	15.533±0.45 ^b	15.533±0.49 ^b	18.732±0.48 ^a	18.732±0.48 ^a

^a For each parameter, values marked with the same letter are not significant for Fischer's LSD at P<0.05.

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8.3 = Carbon isotopic analysis of *Pyrus communis* L. young branches for the comparison of traditional cultivars and new selections

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Pyrus communis L. is a tree species belonging to the family *Rosaceae*, subfamily *Maloideae*. Pear tree is one of the world's oldest cultivated fruit crops. In Europe, it has been cultivated for over 3000 years, with distinct cultivars already documented in 300 B.C. Nowadays the most important Italian region for the cultivation of pear trees is Emilia-Romagna. However, periculture has been facing rising production costs, stagnant market prices, and increasing phytopathological issues for the past five years, undermining the profitability of cultivation. As a result, in Emilia-Romagna the cultivated area shrank from 35,400 hectares in 2011 to approximately 23,700 hectares in 2023. A causal link has been proposed between pear cultivation decline and climate change, also related to a limited genetic diversity, the pear cultivation relying heavily on few traditional cultivars, notably Abate Fétel, which appears highly susceptible to abiotic and biotic environmental stresses (1, 2).

To diversify the pear tree germplasm cultivated in Emilia-Romagna, the University of Ferrara and the Consorzio Italiano Vivaisti (C.I.V.) are collaborating on research focused on soil–rootstock–variety combinations to enhance periculture's resilience to environmental stress. As a first approach to the problem, we focused specifically on 20 varieties/selections of pear tree, including the most spread cultivars for reference (Abate Fétel, Williams); all were planted in an experimental field close to Ferrara in 2023. During the winter resting phase, in December 2024 the youngest branches were sampled and analyzed with respect to the $\delta^{13}\text{C}$ isotopic ratio using Elemental Analyzer–Isotope Ratio Mass Spectrometry (EA-IRMS). The relative amount of ^{13}C isotope in plant tissues primarily depends on the carbon isotopic discrimination by the ribulose-1,5-bisphosphate carboxylase-oxygenase (RuBisCO), which in turn relates primarily to water use efficiency. Therefore, $\delta^{13}\text{C}$ was tested as a reliable proxy for evaluating the plant physiological status based on the organic carbon stored in one-year old branches. It is generally assumed that less negative $\delta^{13}\text{C}$ values characterize more susceptible plants to environmental stressors, particularly water shortage. The results showed a wide $\delta^{13}\text{C}$ variation among cultivars/selections, with values ranging from -28.3‰ to -25.7‰, and, conversely, a small variability among plants belonging to the same cultivar/selection ($\text{CV} < 10\%$, $N=4$). A subgroup of pear selections with $\delta^{13}\text{C}$ of ca. -28‰ is particularly interesting because such values are more typical for leaves than for woody structures, in which $\delta^{13}\text{C}$ usually becomes less negative because of post-photosynthetic effects (3). On one hand, this subgroup should be formed by good, more stress-resistant candidates; on the other hand, the same subgroup includes Abate Fétel, which is instead considered stress-sensitive. This instance suggests that the predictive value of $\delta^{13}\text{C}$ analysis should be enhanced by other independent methods; in this respect, the parameters obtained by chlorophyll *a* fluorescence have also been recorded, and their elaboration is currently in progress.

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8.3 = Phenotypic and Genotypic Characterization of Drought Tolerance in Different Provenances of *Carpinus betulus*

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Climate change is reshaping the composition, structure, and functioning of forest ecosystems by increasing tree mortality rates due to the higher frequency and intensity of extreme events such as droughts and heatwaves. The ability to tolerate water scarcity varies among forest tree species, and potentially among different provenances or genotypes within the same species. We quantified the drought tolerance of different provenances of *Carpinus betulus* through phenotypic trait analysis. Seeds were collected from 11 populations across several Italian regions encompassing a broad range of precipitation and temperature regimes, and were grown under the same conditions in a nursery (Fig. 1). We measured leaf morphological traits (leaf area, specific leaf area, dry matter content) and a physiological trait (turgor loss point, TLP (Fig. 2)) as indicators of drought resistance. These traits were then correlated with climatic conditions at the seed source to identify potential signals of local adaptation, to this end, we performed Pearson correlation analysis to explore linear associations between the TLP and multiple climatic variables. Principal Component Analysis (PCA) was used to summarize the main climatic gradients and reduce dimensionality. Finally, both simple and multiple linear regression models were applied to evaluate the predictive power of climatic variables on TLP. Preliminary results revealed a certain level of variability both between and within populations. Concurrently, high-resolution genotyping using ddRAD-seq was employed to explore the genetic structure of populations and to identify polymorphisms potentially associated with drought tolerance. This multidisciplinary approach provides a robust basis to disentangle the relative roles of phenotypic plasticity and genetic adaptation in shaping drought resilience, and to better predict how forest species may respond to future environmental changes.

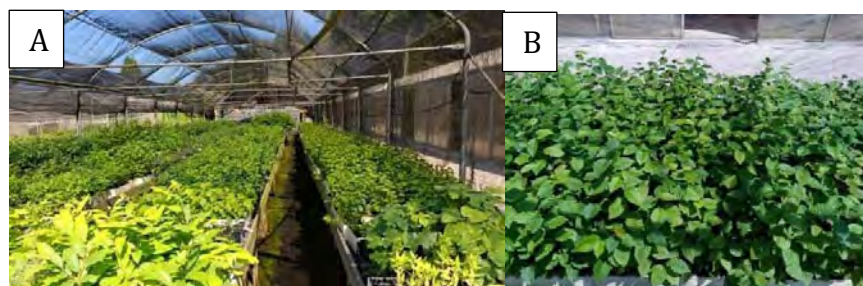


Fig. 1. (A) Overview of the nursery where saplings of *Carpinus betulus* were grown under shade netting with automatic irrigation, ensuring standardized and uniform growth conditions across populations; (B) young saplings of *Carpinus betulus*

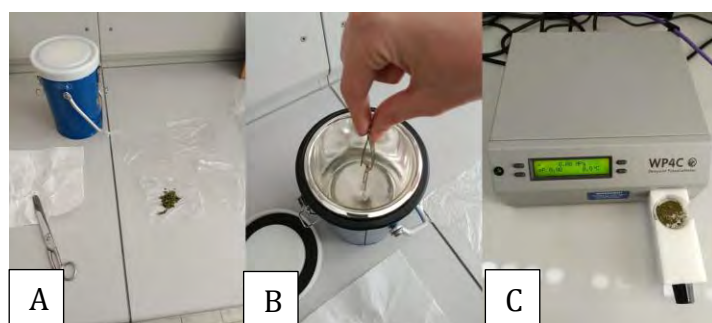


Fig. 2. Workflow for the determination of the Turgor Loss Point (TLP) in leaf tissues. (A) Coarse cutting of leaf material before processing; (B) Leaf sample sealed in plastic film, frozen in liquid nitrogen, and mechanically ground into a fine powder; (C) Processed sample placed in its holder and analysed using a dewpoint hygrometer (WP4) to determine leaf water potential (Petruzzellis *et al.*, 2019) (1)

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9.1 = Subcellular Localization of Mercury Accumulation in Lichen Photobionts: Identifying Cellular Sites of Hg Sequestration

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Mercury (Hg) poses a serious environmental risk due to its widespread contamination and bioaccumulation potential in ecosystems. This potential toxic element (PTE) severely damages photosynthetic organisms by disrupting chloroplast functions, inhibiting photosynthesis, and interfering with cellular metabolism¹. Lichens are known to be excellent bioaccumulators of atmospheric Hg, making them valuable bioindicators of mercury pollution in the environment². However, despite extensive research on lichen-based Hg biomonitoring, there is a significant knowledge gap regarding the exact subcellular locations where Hg accumulation occurs. This ongoing study aims to identify and characterize the specific subcellular compartments responsible for the Hg sequestration in photobionts of *Pseudevernia furfuracea* (L.) Zopf. var. *furfuracea*. This epiphytic lichen is characterized by the presence of isidia, finger-like structures rich in algal cells known to increase the exchange surface with the atmosphere³. Samples of isidiate (IS) and non-isidiate (NI) lichen thalli (Figure 1-2), collected in a pristine subalpine forest (Casera Razzo, Carnic Alps), were exposed in triplicate at eight transplant sites, (Figure 3-5), positioned at the intercardinal points around a waste incinerator in the city of Spilimbergo, Italy, where recent monitoring data confirm ongoing mercury emissions. Following an 8-week long exposure period, samples were retrieved and total mercury content quantified using direct mercury analysis (DMA). Control and exposed lichen lobes were processed for TEM analysis to examine subcellular ultrastructure and identify potential sites of mercury localization with particular focus on chloroplast ultrastructure, specialized organelles and sub-organelles (pyrenoglobules). Surface analysis was used to determine the surface area of isidiate and non isidiate thallus lobes. A comparative analysis between control and mercury-exposed isidiate and non-isidiate samples is currently being conducted to assess their bioaccumulation capacity and the physiological changes associated with mercury exposure.



Fig. 5. Isidiate lobe of *P. furfuracea*



Fig. 3. Exposure devices



Fig. 2. Non isidiate lobe of *P. furfuracea*



Fig. 4. Exposure device in the field

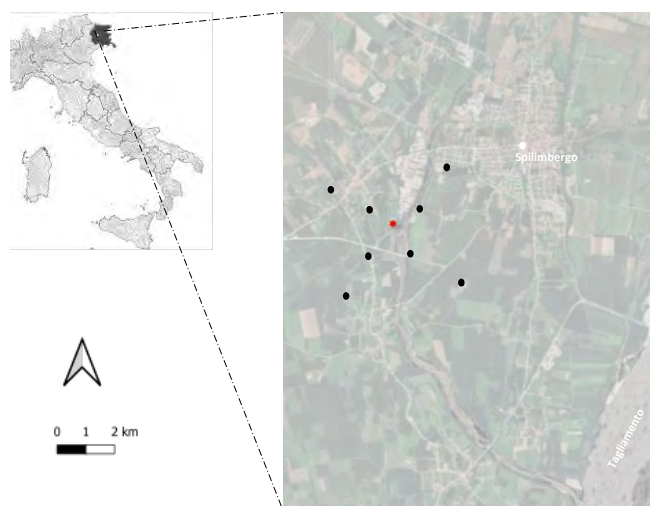


Fig. 5. Localization of the study area with indication of the transplant sites (black dots) and the incinerator plant (red star)

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9.1 = Bioindicators in Forensic science

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The term ‘bioindicator’ encompasses biotic and abiotic responses to ecological change. It includes taxa from multiple Kingdoms used to monitor environmental conditions and detect temporal changes. The bioindicators are deployed in several biotechnological fields, including Forensic, which is the application of the scientific method to crimes. Bioindicators in Forensic science might be used as natural trace as well as for post-mortem intervals. An unexplored area is the Forensic mycology, indeed when it comes to Fungi, very little is known on how these organisms can contribute to solve crimes. Through the awarded project Natural Trace: “Fungi as forensic tools”, funded under the Marie Skłodowska-Curie Actions by the European Commission, our research team will try to contribute to fill the present research gap. The project aims to investigate different aspect of the forensic mycology such as presence/absence, diversity, and growth of fungi in crime scenes as well as forensic important objects. Our model crime scene consists of a garment buried in soil for 1 month and then stored for over a year under controlled environmental conditions to mimic the burial site. The garment served as bait for the fungi. After sampling, potential forensic fungal taxa were isolated axenically, and the growth rate measured. The isolates were also characterized through DNA coding and securely preserved in the culture collection of DISTAV (ColD-UNIGE), which is also a member of MIRRI-IT and project partner of SUS-MIRRI.IT.

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9.2 = Unveiling impacts of *Robinia pseudoacacia* L. and *Prunus serotina* Ehrh. invasions in temperate forest ecosystems of central Europe using multitemporal satellite data

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Biological invasions, one of the main drivers of global change, can cause irreversible shifts in ecosystem composition, structure, and functioning (1). In particular, invasions of non-native trees (NNTs) are causing degradation of forest ecosystems across the entire terrestrial biome. These effects can be both direct (e.g., alteration of soil properties, light intensity, air temperature, or bare soil cover) and indirect (e.g., lower establishment and growth of native plants or reduction of their cover and richness). The complexity of woody invasions requires an integration of monitoring tools at different spatial and temporal scales. Satellite remote sensing, collecting spatial and temporal continuous data, has the potential to contribute to unveil the effects of invasive NNTs (2). In this study, monthly multispectral Sentinel-2 and synthetic aperture radar Sentinel-1 imagery were used to assess the impacts of the two most widespread invasive tree species, *Robinia pseudoacacia* L. (Fabaceae) and *Prunus serotina* Ehrh. (Rosaceae), on productivity, photosynthetic efficiency, and vertical and canopy structures of forests in western Poland. *R. pseudoacacia* and *P. serotina*, both native to North America, were listed in the 100 most invasive non-native species in Europe. They are causing the most severe ecological and socio-economic impacts on European forest ecosystems, particularly sessile oak and Scots pine forests (3). The impacts were analyzed across two forest habitats: nutrient-rich forests dominated by oaks *Quercus robur* L. and *Q. petraea* (Matt.) Liebl. and nutrient-poor forests dominated by Scots pine *Pinus sylvestris* L. We established 160 study plots (0.05 ha), including 64 plots with *R. pseudoacacia*, 64 with *P. serotina*, and 32 (non-invaded) control plots. In each plot, we measured the diameter at the breast height of all trees, calculated the basal area, and estimated the relative dominance of non-native trees. From satellite imagery, a set of spectral metrics were calculated to map the productivity, photosynthetic efficiency, and vertical and canopy structures. The monthly differences between non-invaded and invaded sessile oaks and Scots pine forests were analyzed using linear mixed models (LMMs), one-way ANOVA, and Estimated Marginal Means. The impacts on two forest habitats due to the invasion of *R. pseudoacacia* and *P. serotina* were analyzed along the invasion gradient by LMMs. Results highlighted the effectiveness of the methodology to assess the impacts of *R. pseudoacacia* and *P. serotina* invasion in both habitat forests (conditional R^2 range: 0.795-0.337). Scots pine forest was more sensitive to invasion, with the most intense effects compared sessile oak forest. In Scots pine forest, the impacts were most pronounced in spring and summer, but with opposite trends. Invasion of *R. pseudoacacia* impacted the Scots pine forest in terms of decrease of vertical and canopy structures, causing a closure of the canopy, and incrementing both productivity and photosynthetic efficiency in summer, while it slightly decreased them in spring. On the contrary, the invasion of *P. serotina* was associated with increased productivity in spring and reduced productivity in summer, while both vertical and canopy structures contributed to a greater stratification of the forest architecture. In the oak forest, only the invasion of *R. pseudoacacia* showed impacts. We found a decrease in productivity and photosynthetic efficiency in spring, and an increase in productivity during summer. Thus, multitemporal, multispectral, and radar satellite imagery analysis is an effective tool to assess the context-dependency and temporal variability of non-native invasive tree impacts on ecosystem structure and functioning in different forest ecosystems. This remote sensing approach provides an applied methodological tool easily reproducible and capable of estimating the impacts of NNTs at different levels of invasion, as required by the European Code of Conduct on Planted Forest and Invasive Alien Trees and by the Global Guidelines for the sustainable use of NNTs.

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9.5 = Preserving semi-natural dry grasslands: a cross-disciplinary strategy integrating plant diversity and sustainable land use within the LIFE IMAGINE Project

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Semi-natural dry grasslands are important habitats for plant biodiversity and are protected by the EU 92/43/EEC "Habitats" Directive. Despite their ecological value, these ecosystems are increasingly threatened by the abandonment of traditional grazing practices and, more locally, the intensification of land use, especially in mountainous areas of the central Apennines.

As part of the Integrated LIFE "IMAGINE" Project (LIFE19 IPE/IT/000015), we adopted a cross-disciplinary approach combining vegetation analysis, livestock management planning, and satellite-based monitoring to enhance the management and conservation of these grasslands in the Umbria region.

Based on a comprehensive dataset of both published and unpublished phytosociological relevés, we performed a floristic, vegetational and ecological characterization of the different grassland types, mainly referable to Annex I habitats 6210 "Semi-natural dry grasslands and scrubland facies on calcareous substrates (*Festuco-Brometalia*)" and 6230* "Species-rich *Nardus* grasslands, on siliceous substrates in mountain areas".

To determine the appropriate grazing pressure for each grassland type, the Pastoral Value (PV) was calculated. These results guided the development of "Biodiversity-focused" Grazing Plans, including rotational grazing systems to ensure sustainable pasture use (Action C14).

Fieldwork further provided detailed insight into grassland pressures and threats, supporting the elaboration and implementation of site-specific Action Plans (Action C15). These included conservation actions such as shrub clearing to counteract encroachment processes, and the eradication of invasive plant species, both native (e.g. *Asphodelus macrocarpus* Parl.) and alien (e.g. *Senecio inaequidens* DC.).

Additionally, a Web-GIS application was developed using Sentinel-2 imagery and harmonic modeling of the Enhanced Vegetation Index (EVI) within Google Earth Engine. This tool allows for real-time monitoring of grassland growth and supports informed grazing management.

This integrated strategy – grounded on vegetational and ecological knowledge of grassland areas, adaptive livestock planning, and adequate technological tools – aims to promote a conscious and more sustainable grassland management. Furthermore, the LIFE IMAGINE Project highlights the fundamental role of farmers in the active conservation of biodiversity.

9.5 = Restoration of Annex I Habitats Carried Out within the LIFE IMAGINE Project (LIFE19 IPE/IT/000015 “Integrated Management and Grant Investments for the Natura 2000 Network in Umbria”)

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The LIFE IMAGINE project (<https://www.lifeimagine.eu/>) aims to implement a unified, coordinated, and participatory strategy for integrated management in Umbria in order to achieve the conservation objectives set forth by the Habitats and Birds Directives, based on the content of the Prioritized Action Framework (PAF). The project contributes to the broader goals of regional development.

Among the concrete actions foreseen by the project are interventions for the conservation and restoration of rare habitats associated with wetland ecosystems, which are only found at a few locations within the Umbria Region. For these habitats, once the conservation status was assessed (through analysis of structure and function parameters, and identification of pressures and threats), concrete restoration and recovery actions were carried out as follows:

- **Habitat 7210 Calcareous fens with *Cladium mariscus* and species of the *Caricion davallianae****. This habitat is present in the Umbria Region only along the shores of Lake Piediluco (TR) (SCI IT5220018 and SPA IT5220026) (1). Activities included mapping all known occurrences of the habitat, assessing conservation status, and identifying potential pressure factors. In suitable areas, actions were implemented to enlarge the habitat by at least 1,000 m². Restoration involved planting individuals of *Cladium mariscus* (L.) Pohl, propagated from locally collected seeds, in association with *Carex pseudocyperus* L. at a 4:1 ratio. These were planted both along the shoreline and on specially designed artificial floating islands (AFIs) to reconstruct the floating vegetation mats.

- **Habitat 7230 Alkaline fens**. In Umbria, this habitat is limited to the Colfiorito Marsh and the Monti Sibillini National Park area (SCI/SPA IT5210072 and IT5210071). This is a high-value habitat subject to natural succession in the absence of appropriate hydrological conditions or active management. Traditional practices, such as mowing or cutting woody species, have played a crucial role in maintaining the habitat. Conversely, the abandonment of these practices in recent decades has led to a significant decline in habitat extent, especially in the Colfiorito Marsh. Once the current extent and conservation status of the habitat were clearly defined, the following actions were undertaken: a) Control of secondary successional species resulting from mowing abandonment; b) *In situ* seed collection and *ex situ* germination/propagation of typical habitat species (2,3) i.e. *Eriophorum latifolium* Hoppe, *Carex panicea* L., *Epipactis palustris* (L.) Crantz, *Anacamptis laxiflora* (Lam.) R.M.Bateman, Pridgeon & M.W.Chase, etc.; 3) Establishment of a nursery for future transplanting phases.

- **Habitat 92A0 Riparian galleries of *Salix alba* and *Populus alba***. These habitats generally exhibit an "unfavourable-bad" conservation status and therefore require active management interventions. In particular, along the Tiber River in Umbria, plant communities show signs of degradation due to indiscriminate cutting, which has caused severe fragmentation of the habitat and facilitated the spread of invasive alien species, most notably *Robinia pseudoacacia* L. To counter the decline in phytocoenotic quality, interventions were planned to mechanically control *Robinia pseudoacacia* L. over approximately 10 ha, accompanied by the planting of typical habitat species aimed at improving forest structure and floristic composition (structure and function parameters).

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9.6 = From field to satellite: linking plant functional traits to greening trends through vegetation resurveys

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Resurvey of vegetation plots is an invaluable way of studying long-term vegetation dynamics. Resurvey data inform us about spatio-temporal changes in plant species composition, cover, richness and functional composition following environmental changes (e.g. colonisation, land use change, climate change). With a growing concern about the impact of global change on ecosystems and natural habitats, historical plots have been increasingly resurveyed in the past decades. In parallel with resurvey studies, there has been a widespread diffusion of remote sensing-based research on ecosystem processes in mountain areas. The European Alps have been extensively studied, and of all the processes, the greening of alpine vegetation – typically measured through increases in the Normalized Difference Vegetation Index (NDVI) – has attracted the most interest. Most studies point to the relationship of increasing NDVI with climate related factors like warming, altered precipitation patterns and reduced snow cover, and with site elevation and topography. Furthermore, greening trends have been related to sparsely vegetated areas such as screes, cliffs and glacier forefields; areas with increasing shrub cover near treelines and areas where there is a long-lasting snow cover. Overall, to fill the gap between greening trends and underlying ecological processes it is needed to relate the remote sensing findings with ground-truth vegetation data

The aim of this study is to use, for the first time as far as we know, resurvey data from vegetation plots to explain the observed greening trends across the Alps. Given that different environmental processes drive increases in primary production with varying intensity across vegetation types and successional stages, we believe that analysing long-term vegetation changes in sampled communities can provide valuable insights into these trends. Vegetation changes linked to productivity shifts can be assessed through variations in plant cover, functional traits related to the “fast-slow” leaf economics spectrum, and life-form assemblages. We hypothesize that these variables may be more important in explaining greening trends than environmental variables alone. Within this framework, we expect distinct trajectories of vegetation change under greening trends depending on habitat type. We thus additionally hypothesize that: (1) In sparsely vegetated areas, greening results from an overall increase in plant cover; (2) In grasslands, by contrast, greening is driven by the arrival of more competitive or taller species replacing small, stress-tolerant ones; (3) An increase in chamaephytes and phanerophytes further contributes to the greening trends in advanced stages of grassland succession.

Overall, the considered biological variables have a similar importance compared to the environmental ones in explaining the absolute measured productivity. Among them, leaf area, plant height, and total plant cover are the most important, showing a clear relation with NDVI. However, only the plant height shows a significant effect on the increasing NDVI values across surveys when considering longer time series. An analysis of shorter time series – for a more cautious approach – reveals no effect of either biological or environmental variables across surveys. Furthermore, the distinction of alpine communities into habitat type did not show different relations between traits, life forms, and plant cover with NDVI across time. Increasing values of NDVI are directly related to increasing values of plant height, leaf area, and plant cover for all the studied habitats. Habitat only acts as a factor increasing productivity along the vegetation succession due to a corresponding increase in plant cover and traits related to competitive life strategies.

9.6 = A National Baseline for Biodiversity: Systematic Monitoring of Annex I Habitats in Italian National Parks under the DigitAP Framework

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Habitats, shaped by complex biotic and abiotic interactions, are fundamental components of biodiversity and vital indicators of conservation status. The EU Habitats Directive (92/43/EEC) mandates Member States to monitor Annex I habitats (Art. 11) to track conservation trends and assess policy effectiveness. However, standardized monitoring approaches remain under development, while biodiversity loss continues to accelerate. Supported by the National Recovery and Resilience Plan (NRRP), Italy is implementing its first national-scale habitat monitoring program within Protected Areas, under the DigitAP project—promoted by the Ministry of the Environment and Energy Security (MASE) and coordinated by ISPRA (1). The key goal of DigitAP is monitoring Annex I habitats across Italian National Parks through vegetation surveys aimed at establishing a permanent observation network for medium- to long-term assessments. A specialized team of surveyors, coordinated by a Temporary Grouping of Companies and supervised by the Italian Botanical Society (SBI), was engaged to carry out two years of annual vegetation surveys. The project targets 98 Annex I habitats across 1,857 stratified-random 100×100 m² cells. Data are collected in accordance with national guidelines (2) and validated by habitat- and region-specific experts to ensure quality, accuracy, and comparability, with photographic and video documentation supporting assessments. Sampling design is based on Annex I habitat maps in each park, with cell allocation proportional to habitat extent and timing aligned with habitat type (3) and phenological stage. Standardized sampling areas per habitat type have been defined. Ground-truthing, a pivotal phase, requires close collaboration among field teams, validators, and coordinators. Custom GIS-based applications support fieldwork and data management, while a centralized server enables real-time storage, visualization, periodic backups, and interactive dashboards for quality assurance. Multimedia archives facilitate remote validation. In the first year, initial activities focused on defining permanent plots and documenting key structural species and environmental features. The 2025 campaign is gathering full floristic-vegetation data, forming a baseline for long-term monitoring. Data will feed into the Italian National Biodiversity Network (NNB). This effort marks the first coordinated national initiative for Annex I habitat monitoring in protected areas, representing a crucial step toward building a harmonized, effective conservation system. The initiative advances the implementation of the Habitats Directive in Italy and Europe, supporting long-term biodiversity protection and reaffirming the value of traditional fieldwork in ecological monitoring.

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9.6 = A transferable machine learning model for Alpine peatland mapping using multi-source remote sensing images

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Peatlands provide crucial ecosystem services, including carbon storage, biodiversity support, and climate regulation. However, they are increasingly threatened by climate and land-use changes, which accelerate carbon release and degradation. At the southern edge of their distribution, peatlands are rare, fragmented, and often underrepresented in existing maps. In particular, in alpine regions like the Italian Alps, the inventories are often outdated and incomplete. Building on a previous model developed for the Eastern Alps, this study tested its applicability to the Western Alps, where peatlands are generally rarer and have different environmental conditions. Using a combination of multi-source remote sensing imagery (i.e.

Sentinel-1 and 2, Digital Elevation model) and a tailored mask, we trained the model on 70% of digitised peatlands and validated it on the remaining 30%. We also tried to improve the model thanks to vegetation classification data. The results confirm that the model is transferable across the alpine region, and if based on vegetation data, it yields improved accuracy. This approach offers a promising tool for large-scale peatland mapping and provides a valuable basis for targeted conservation strategies.

9.6 = Study of *Cymodocea nodosa* meadow and blue-carbon stock, is there a correlation with seagrass cover

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Seagrass meadows play a pivotal role in coastal carbon dynamics through the sequestration of organic carbon in their underlying sediments, a component of what is termed "blue carbon" (Nellemann et al., 2009). While extensive research has focused on *Posidonia oceanica*, less attention has been paid to minor seagrass species such as *Cymodocea nodosa*, particularly in the Mediterranean region (Waycott et al., 2009). This study aims to investigate the relationship between *C. nodosa* coverage and sedimentary organic carbon storage along the Italian coastline. The research integrates ecological and geological analyses, including sediment granulometry and soil type classification, to evaluate the capacity of *C. nodosa* meadows to trap sediments and sequester organic carbon.

Given the scarcity of region-specific conversion indices for estimating organic carbon from more accessible proxies, a key objective of this project is the development of a Mediterranean-specific conversion index linking Loss on Ignition (LOI) measurements to organic carbon concentrations obtained via Elemental Analyzer (CHN). This index is conceptually analogous to that of Fourqurean et al. (2012), developed for Floridian seagrasses, but adapted for *C. nodosa* habitats. Seagrass coverage is quantified using image analysis software (ImageJ) on photographic data of quadrats from which sediment cores are extracted, enabling a robust correlation analysis between vegetative cover and organic carbon stocks. The ultimate goal of this research is to quantify and validate the ecosystem service provided by *C. nodosa* in sediment stabilization and carbon sequestration, thereby contributing critical data for the inclusion of this species in future blue carbon assessments and conservation strategies.



Fig. 1a-1b-1c. *C. nodosa* meadows in Nora Lagoon (CA) Italy, *C. nodosa* Herbaria specimen and *C. nodosa* meadows in Sant'Antioco Lagoon (SU) Italy

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9.6 = Photovoltaic Systems and Plant Biodiversity in Sicily

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In recent decades, driven by the need to reduce greenhouse gas emissions and contrast climate change, the realization of photovoltaic (PV) has become a widespread practice. However, the conversion of agricultural or natural land into sites for solar energy production raises concerns about environmental impacts, particularly on local flora and soil quality (1). Conversely, PV installations can have some environmental benefits. For example, the absence of agricultural activity in these areas can reduce the use of pesticides and herbicides, decreasing soil contamination and potentially promoting the spread of certain plant species (2). This requires an assessment of both negative impacts and potential benefits. Currently, scientific literature presents significant gaps regarding the long-term effects of PV systems on flora, plant communities and soil quality (3). There is an urgent need for detailed studies, especially considering the significant increase in PV park installations in recent years, particularly in central-southern Sicily. This area is highly suited for such installations due to climatic, socio-economic and topographical reasons. This study aims to analyze the impact of land-based photovoltaic installations on flora, vegetation and soil quality. Through field studies and comparative statistical analyses, the effects on plant biodiversity, changes in floristic composition, and microclimatic effects will be evaluated. Additionally, the analysis of plant communities will be deepened, with the goal of updating territorial data and enhancing the knowledge base of the area. This will include integrating ecological indices designed to assess the environmental suitability of sites intended for PV installations, fostering a multi-criteria approach to energy planning. Finally, another objective of the study is to propose mitigation measures based on the results obtained, to reduce the negative impact on local flora, in order to minimize the negative effects on biodiversity and improve soil quality. Preliminary results indicate that areas designated for PV installations are often agricultural lands characterized by severely impoverished flora due to intensive farming practices (Fig. 1). The compensation and mitigation measures currently in place are frequently generic and fail to account for local ecological characteristics, highlighting the need for more targeted and informed strategies.



Fig. 1. Area targeted for the installation of the photovoltaic system. Mazzarino territory

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9.7 = Impact of Global Changes on the Vegetation in a Protected Area: A Twenty-Year Analysis in Aspromonte National Park

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Global changes represent one of the most pressing threats to ecosystems, profoundly influencing habitats and redefining management and conservation priorities. Rising temperatures, altered precipitation regimes, alien species and the increasing frequency of extreme events, such as prolonged droughts and wildfires, are modifying the composition, structure, and resilience of forests (1). Often, these changes result in habitat fragmentation, which isolates populations and diminishes their ability to adapt. This situation calls for an urgent reassessment of traditional protected area management practices. It is essential to emphasize conservation strategies that prioritize adaptation and the maintenance of biodiversity in response to a changing climate, also contrasting the new entry and the diffusion of alien species. For this reason, this study aims to analyze the impact of global changes on forest vegetation within protected areas, using Aspromonte National Park, a highly biodiverse region, as a case study. It evaluates the transformations in habitat cover and fragmentation over twenty years by comparing the 2002 vegetation map of Aspromonte National Park (2) and the Nature Map of the Calabria region (3), to quantify spatial and temporal habitats variations using QGIS software. The Morphological Spatial Pattern Analysis (MSPA) and FRAGSTATS v4.2 were employed to evaluate habitat fragmentation.

Preliminary results indicate that forest habitats had a different trend. *Quercus frainetto* forests and *Quercus petraea* subsp. *austrotyrrhenica* forests (habitat 91M0) and *Quercus pubescens* forests (habitat 91AA*) show signs of vulnerability and potential decline, alongside increased fragmentation. *Fagus sylvatica* forests (habitat 9210*) now occupy a larger area than in the past, likely due to more effective protection provided by the park. *Pinus nigra* subsp. *laricio* forests (habitat 9530*) exhibit a significant reduction in occupied areas, which is probably due to repeated fires in previous years. A slight decrease was also observed for *Quercus ilex* forests (habitat 9340).

The analysis emphasises the need for proactive conservation strategies that consider forests' intrinsic adaptive capacity and support a shift from an exploitation-based management model to one focused on protection and ecological restoration, while counteracting habitat fragmentation and the entry and diffusion of alien species. In conclusion, this work provides a scientific basis to guide conservation policies and promote the resilience of autochthonous forest communities in the face of global changes, ensuring their survival for future generations.

1) J. Tollefson, IPCC climate report: Earth is warmer than it's been in 125,000 years. *Nature*, 596 (2021), pp. 171-172

2) G. Spampinato, P. Cameriere, D. Caridi, A. Crisafulli (2009) Carta della biodiversità vegetale del Parco Nazionale dell'Aspromonte (Southern Italy). *Quaderno di Botanica Ambientale Applicata* 2009, 20, 3–36

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This work is financed by the Research Project “Eradication of crimson fountaingrass in Calabria: Prevention, eradication and study of the potential expansion of its habitat” - CUP: C37G24000480002, funded by the Region of Calabria (Italy).

9.9 = Moss-based phytoremediation in urban settings: evaluating particulate matter capture by three moss species

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Air pollution, particularly airborne particulate matter (PM), represents a major threat to human and environmental health in urban areas. While green infrastructure is increasingly adopted for atmospheric remediation, conventional vertical greening systems are often hindered by high maintenance costs and complex management.

In this context, Bryophytes—due to their structural simplicity, resilience, and capacity to accumulate airborne pollutants—emerge as effective agents for low-maintenance phytoremediation.

This study assesses the PM capture efficiency of three moss species: *Barbula unguiculata* Hedw. (Pottiaceae), *Grimmia pulvinata* (Hedw.) Sm. (Grimmiaceae), and *Homalothecium sericeum* (Hedw.) Bruch, Schimp. & W. Gümbel (Brachytheciaceae). These species were integrated into a modular moss-based system (MosSkin) designed for building envelope applications.

The system was deployed for three months in a high-traffic urban location. After exposure, moss phyllids were analyzed using Field Emission Scanning Electron Microscopy (FESEM) to quantify particle density, size distribution, and composition across three leaf zones (base, middle, apex).

Results showed particle densities up to 45,000 particles per mm², with *H. sericeum* and *G. pulvinata* demonstrating superior capture capacity. *G. pulvinata* showed selective affinity for particles between 2–7.5 µm, and all species retained a substantial fraction (45–55%) of ultrafine particles (PM_{0.5}). Data were compared with ARPAL air quality monitoring station to evaluate the potential application of such results and to obtain the real particle capture ability. Differences in retention efficiency were linked to species-specific morphological traits and microstructural characteristics of phyllids.

These findings highlight the phytoremediation potential of moss-based systems in mitigating urban air pollution. The MosSkin approach provides a scalable and low-input solution that complements existing green infrastructure strategies. Further investigation under variable environmental stressors could elucidate the mechanistic basis of PM capture, supporting targeted application of mosses in phytoremediation of urban atmosphere.

9.9 = Exploring phytoremediation potential for nitrocellulose-contaminated soil: a case study from Spilamberto (Modena)

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The study investigates the effects of a specific soil pollutant on seed germination and plant growth, with the aim of exploring the potential of phytoremediation as a strategy to address and resolve an environmental issue. Phytoremediation utilizes the natural ability of plants to take up, accumulate, degrade, or stabilize pollutants from soil, water, and air, and it is considered an environmentally sustainable and cost-effective technique for the remediation of contaminated sites. In this context, the study presents a case in which plants could play a key role in addressing a specific environmental problem located in the Municipality of Spilamberto (Modena). Here, historical explosive manufacturing activities have led to the contamination of a large area (approximately 50 hectares) with nitrocellulose (NC). As part of a broader strategy aimed at transforming waste into a resource (End of Waste), the project seeks to explore the accelerated biodegradation of NC in soil, promoting the release of nitrate ions and evaluating their potential use as fertilizing agents. To assess the feasibility of using plants as a strategy for managing NC-contaminated soil, the project initially aimed to investigate the effects of NC on seed germination and plant growth.

The phytotoxicity of NC-contaminated soil was assessed through germination bioassays using *Lepidium sativum* L., while the effects on plant growth were evaluated with *Lactuca sativa* L. and *Triticum aestivum* L., representing dicots and monocots respectively. Three substrate concentrations were tested: 20% NC (w/w), obtained by mixing NC-contaminated soil (34% NC w/w) with standard substrate, and two additional treatments where NC had been partially degraded by *Nitrosomonas* sp., resulting in final concentrations of 12% (NB1) and 10% (NB2). Germination tests followed APAT guidelines, assessing seed germination (SG) and root elongation (RE), with results expressed as Germination Index (GI%). Growth tests were carried out in pots under controlled conditions.

Soils containing untreated NC (20%) showed high GI values (>60%), indicating no phytotoxicity. In contrast, soils with bacterially treated NC (NB1 and NB2) showed significantly reduced GI values, particularly at higher concentrations.

Interestingly, despite the reduced germination, substrates with bacterially digested NC had a positive effect on plant growth. Agronomic (biomass, allometric measures, LAI) and physiological (Dualox) analyses revealed a slight but significant ($p < 0.05$) increase in plant growth on NB1 substrates. These findings suggest that while bacterially treated NC may initially exhibit phytotoxic effects—possibly due to nitrate accumulation—its degradation products may later act as fertilizers, enhancing plant growth under certain conditions.

This study highlights the complex interaction between NC degradation products and plant development, suggesting that, the transformation of NC-contaminated soil through microbial treatment may enhance plant growth, supporting the feasibility of phytoremediation as a strategy to manage a specific soil contamination issue.

9.10 = Restoration Ecology and Urban Biodiversity: The SERRA Project Approach

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The SERRA project (Specie Erbacee tolleranti per la Rigenerazione urbana, meaning Tolerant Herbaceous Species for Urban Regeneration) aims to support the urban ecological transition through the selection, production, and use of native herbaceous species from the Italian Mediterranean flora, suitable for the rehabilitation of marginal and degraded urban areas. The primary objective is the development of seed mixtures tailored for restoration ecology interventions in environments exposed to biotic and abiotic stressors, such as high temperatures linked to urban heat islands, high relative humidity, and soil pollution. The selected species are chosen not only for their adaptive capacity and tolerance to contaminants such as heavy metals, but also for the ecological functions they provide, including enhancing urban biodiversity, supporting pollinating insects, and activating regulatory ecosystem services. SERRA directly aligns with the objectives of Spoke 5 of the National Biodiversity Future Center (NBFC), promoting innovative and integrated green solutions for sustainable urban regeneration.

The project follows an integrated research and experimental development pathway structured in multiple phases: selection of Mediterranean herbaceous species, assessment of their tolerance to contaminants, analysis of their interactions with pollinators, and study of plant phenology and development within urban environments. The expected outcomes include the creation of suitable species lists and seed mixtures to be used in ecological urban interventions through a direct interaction with local garden nursery production chains. Following the species selection phase, the project has moved into operational implementation. In collaboration with the Municipality of Cagliari, specific sites have been identified for planting the selected species, in order to concretely apply the developed solutions in sustainable urban regeneration.

SERRA does not merely deliver an innovative product, namely the selected seed mixture, but also aims to generate applicable and transferable knowledge capable of stimulating the gardening nursery sector, strengthening local production chains, and promoting a systemic approach to urban green space management. The integration of native herbaceous species into urban greening programs not only enhances the ecological resilience of cities but also significantly contributes to the creation of functional landscapes that provide multiple benefits for biodiversity and human well-being. In a context of increasing anthropogenic pressure and climate change, the expected results of the project represent an essential scientific and practical contribution toward an effective, replicable, and sustainable ecological transition.

9.10 = Hourly readings: valuable tool for preventing allergic attacks?

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Despite the continuous increase in allergies, aerobiological monitoring as a useful research tool for the prevention of these diseases has not yet been made mandatory.

One aspect of monitoring that has proven decisive in terms of prophylaxis is the relevance of its year-round application for allergic patients to limit exposure to pollen sources that cause pollinosis.

The aim of this research is to investigate the pollen fluctuations of the allergenic flora and in particular to study the hourly trend in concentrations at a specific time of day and to evaluate the trend over the entire day during the hours of maximum pollen concentration using the *Intradiurnal Index* (IDI) (1) of nine herbaceous and arboreal *taxa*, chosen on the basis of their major relevance from an allergological point of view and representative nature of the flora in Rome.

The analysis of the pollen data collected covers an entire calendar year (2010) for a total of 365 slides from the “San Pietro” Fatebenefratelli (RM5) monitoring station, located in the northernmost part of the city and annexed to the Aerobiological Monitoring Centre of Rome Tor Vergata. The choice of the station and the reference year was not left to chance, as 2010 was the year with the highest number of daily samplings for this urban-type station and, as such, more exposed to vehicular traffic, the cause of allergic symptoms and related exacerbations, than rural areas. Knowing the intradiurnal fluctuations of pollen is useful for comparison with pollutants such as NO₂ and O₃ (Fig. 1) which are considered particularly harmful to public health.

The hourly readings (24 intervals of 2 mm each on the 48 mm slide) for the quantitative and qualitative analyses of the hourly data of the *taxa* were performed following the guidelines suggested by the standardised routine methodology used (2).

The tree *taxa* Cupressaceae/Taxaceae, *Alnus* sp., *Betula pendula* Roth., *Carpinus betulus* L./ *Ostrya carpinifolia* Scop., *Corylus avellana* L., *Castanea sativa* Mill., *Quercus* sp., *Fraxinus* sp., *Olea europaea* L., *Populus* sp. and the herbaceous *taxa* Poaceae, Urticaceae showed two completely different types of intradiurnal trends:

1. most *taxa* showed a bell-shaped pattern (lower pollen concentrations in the early hours of the day, a peak in the warmer afternoon hours and a decrease in concentrations in the evening hours) subject to the influence of meteorological variables (temperature and precipitation) (Fig. 2).

2. a small number of species displayed a rather homogeneous trend throughout the day.

This work of analysis has further highlighted the need for, and importance of, limiting the exposure of atopic and allergic patients in the urban air of Rome and elsewhere to pollen sources as much as possible at certain times of the day, in order to reduce allergic attacks and, in the most severe cases, asthmatic attacks.

It is therefore evident that it is necessary to make aerobiological monitoring mandatory in order to investigate and collect information on the daily and hourly trend of allergenic species, allowing medical assistance with predictive, prognostic and appropriate immunotherapeutic diagnoses and therapies as specific as possible for allergic users. This would promote greater awareness among allergy sufferers and treating physicians, enabling an overall assessment of each patient's pathogenesis.

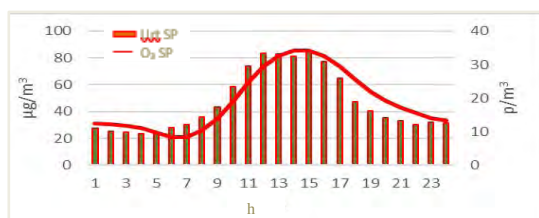


Fig. 1. Comparison of hourly trends in Urticaceae and O₃ at station RM5

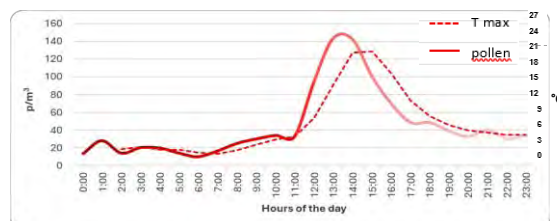


Fig. 2. Example of hourly trend of Poaceae and T max at station RM5

1) CEN/TS 16868:2019. Ambient Air - Sampling and Analysis of Airborne Pollen Grains and Fungal Spores for Networks Related to Allergy – Volumetric Hirst Method

2) M.M.Trigo, M. Recio, F.J. Toro, B. Cabezudo (1997). Intradiurnal fluctuations in airborne pollen in Málaga (S. Spain): A quantitative method. Grana, 36, 39–43

9.10 = Aerobiological monitoring for the good use of green spaces and the promotion of health and well-being

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Most of the world's population lives in urban areas and this growing trend is expected to continue in the coming years. Nevertheless, city dwellers are particularly exposed to air pollution and global warming effects. In this context, pollinosis is increasing and allergic diseases, such as asthma and rhinitis, affect up to 30% of the inhabitants of industrialized countries (1).

In order to implement sustainable and health-oriented urban planning policies, *Nature-based solutions* and, in general, so-called ecosystem services, i.e. those benefits that natural ecosystems provide to humanity, are recently becoming more widespread. Among these, the promotion of green areas and urban forests (2) play a leading role.

However, the correct promotion of urban green spaces cannot be separated from the knowledge of the adverse effects that some botanical species can have on sensitive subjects.

For this purpose, aerobiological monitoring, that studies the presence and concentration of airborne biological particles (such as pollen, fungal spores, etc.) can represent a key tool for assessing environmental quality and its impact on public health, especially in urban and peri-urban green spaces.

This work is carried out within the research project “The good use of green and blue spaces for the promotion of health and well-being”, in short VeBS, implemented with the technical and financial support of the Italian Ministry of Health within the National Investment Plan Complementary to the PNRR (PNC) – Investment E.1 Health-environment-biodiversity-climate.

The project aims to promote the correct use of urban green and blue areas and to implement policies for planning, maintenance, management to improve knowledge of the effects associated with health. An atlas of the main plant species will be created based on their habitat, their ability to counteract the effects of human activities, to provide ecosystem services identified as relevant for the urban environment, while minimizing the negative impacts of green infrastructures on human health (allergenic pollens) and ecosystems (invasive alien species). In addition, VeBS project focuses on case studies in selected areas to provide useful knowledge for optimal management and use of green infrastructures.

For the city of Rome, concentration trends of the main allergenic families were calculated using the seasonal Kendall test for a period of 17 years (3). In Fig. 1 the trend of Cupressaceae and Poaceae pollen is shown.

The analysis of historical series of aerobiological data allows to evaluate the trends of allergenic pollen concentration (and consequently the diffusion of the corresponding species) so as to improve the management of public green and provide useful information to citizens for the good use of green spaces.

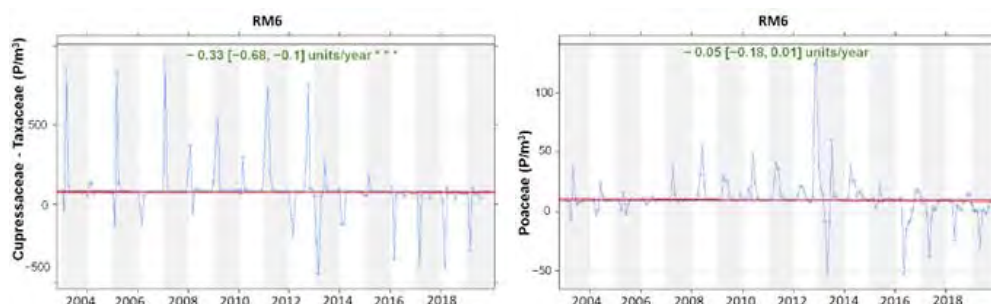


Fig. 1. Kendall trend test results for Cupressaceae and Poaceae pollen in the RM6 station for the period 2003–2019

1) EAACI, Global Atlas of Allergy, European Academy of Allergy and Clinical Immunology. 2014, www.eaaci.org.

2) FAO, FAO. 2016. Guidelines on urban and peri-urban forestry, by F. Salbitano, S. Borelli, M. Conigliaro and Y. Chen. FAO Forestry Paper No. 178. Rome, Food and Agriculture Organization of the United Nations.

3) A. Di Menno di Bucchianico, R. Gaddi, M.A. Brighetti, D. De Franco, A. Miraglia, A. Travaglini (2023) Sustainability 15, 6150, <https://doi.org/10.3390/su15076150>

9.10 = Influence of water stress on urban plant efficiency in capturing particulate matter: a case study with *Loropetalum chinense* var. *rubrum*

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Urban vegetation plays a crucial role in mitigating air pollution by intercepting airborne particulate matter (PM) on leaf surfaces. Among the various environmental stressors affecting plant performance, water stress is particularly relevant in urban contexts, where irregular irrigation and extreme microclimates can alter leaf morphology and particulate deposition dynamics.

This study investigates the ability of *Loropetalum chinense* var. *rubrum*, an ornamental evergreen shrub, to retain PM under conditions of differential water availability, simulating drought stress conditions commonly found in Mediterranean cities. A key feature of this research was the active involvement of citizens. The selection of 16 balconies across the city of Turin was guided by an online questionnaire distributed to a group of resident volunteers. On each balcony, two *Loropetalum* plants were placed: one regularly irrigated (wet) and the other subjected to water stress (dry). This participatory approach allowed the collection of essential micro-environmental data to identify the most suitable sites for the experiment.

Drawing inspiration from the methodology of Ristorini et al. (2020), which emphasizes the use of vacuum filtration and chemical fractionation to separate and characterize the water-soluble and insoluble PM fractions on leaf surfaces, we quantified PM deposition and heavy metal accumulation on leaves exposed to different microclimatic and watering regimes on urban balconies. Leaves were washed, and the collected solutions were filtered and analyzed gravimetrically and chemically.

Significant differences were found in total PM accumulation and metal content between drought-stressed and well-watered plants. Results show a significant reduction in total particulate matter deposited on leaf surfaces in dry-stressed plants (19.0 ± 5.3 mg) compared to well-watered controls (34.4 ± 14.2 mg). The chemical composition of the particulate analyzed by ICP-MS differed between treatments. Both the total ionic concentrations in the washing solutions and the concentrations of heavy metals retained on the filters after digestion revealed distinct profiles suggesting a strong influence of water availability on the capacity of leaves to capture and retain airborne pollutants.

Our findings highlight the importance of physiological and morphological leaf traits influenced by water stress in modulating PM capture efficiency, with implications for species selection in urban greening strategies. The study also demonstrates the value of integrating citizen science, nature-based solutions, and advanced analytical techniques to enhance air quality monitoring in urban environments.

1) M. Ristorini, C. Baldacchini, L. Massimi, G. Sgrigna, C. Calfapietra, (2020). International Journal of Environmental Research and Public Health, 17(16), 5717

9.10 = Plants rhizosphere impact on microbial biodiversity and antibiotic resistance in an urban soil in Milan, Italy

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Urban soil pollution is an emerging environmental issue with significant implications for ecosystem health and public safety. Among the various pollutants, antibiotics, recently considered emerging contaminants, are of particular concern due to their potential to disrupt soil microbial communities and promote the spread of antibiotic resistance (AR). In this context, Nature-Based Solutions (NBS), involving the synergistic use of plants and their associated microbial communities, offer a sustainable remediation strategy.

In this preliminary study, we investigated microbial biodiversity in an urban brownfield located near a disused railway yard in Milan, Italy.

Bulk soil and local plant rhizosphere samples were collected from fifteen representative points across the site. The number of culturable bacteria was estimated via plate counts and taxonomically identified using the hypervariable regions of the 16S rRNA gene sequencing, complemented by Next Generation Sequencing (NGS).

Approximately 300 bacterial strains were isolated and characterized at both phenotypic and molecular levels. Gram staining revealed a predominance of Gram-negative bacteria (70%) over Gram-positive (30%), with morphological diversity including bacilli (72.8%), cocci (6.0%), coccobacilli (15.5%) and filamentous forms (5.7%). Microbial abundance was markedly higher in plant rhizosphere samples compared to bulk soil. In contrast, the Shannon and Simpson biodiversity indices, calculated from both NGS data and culturable isolates, indicated greater microbial diversity in bulk soil than in the rhizosphere, reflecting the selective pressure exerted by root exudates that may favor specific microbial taxa.

All isolates were screened for resistance to five antibiotics (Ampicillin, Tetracycline, Rifampicin, Gentamicin and Vancomycin) across a concentration gradient (0.2 $\mu\text{g mL}^{-1}$ to 10,000 $\mu\text{g mL}^{-1}$). Survival rates, monitored at 28°C after 24/48 hours, confirmed the widespread presence of antibiotic-resistant bacteria, with 56.8% of isolates exhibiting high levels of resistance. In fact, complete bacterial mortality was observed only for Vancomycin at the high concentration of 3,000 $\mu\text{g mL}^{-1}$.

These highly resistant strains were less frequent in the areas of the field with the presence of dense vegetation (trees and bushes), suggesting that some environmental factors, such as root exudate and microbial interactions, may influence AR distribution and persistence.

Our research provides preliminary insights into microbial dynamics and antibiotic resistance in urban soils. It emphasizes the ecological importance of rhizosphere processes and supports the development of Nature-Based strategies for urban soil restoration, aimed at enhancing microbial and plant biodiversity, mitigating contamination, and promoting overall soil health.

9.10 = Plant cover as an indicator of soil and vegetation features in alpine limestone screes: a case study in the Italian Alps

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Alpine limestone screes represent highly dynamic and fragile ecosystems, where vegetation establishment and soil development occur under extreme environmental constraints. This study explores the relationships between plant cover, plant community composition, and soil characteristics during the natural colonization process of limestone screes in an area of the Italian Alps (Mt. Cavallo, Orobie Alps). A total of 50 plots were surveyed along a gradient of increasing plant cover. For each plot, both floristic-vegetational and soil parameters - including plant cover, number of species, soil depth, texture, pH, organic matter, and total nitrogen - were recorded. The results reveal a clear pattern of ecological succession, with early-stage screes exhibiting low plant cover, poor soil development, and limited species richness, dominated by pioneer species of *Thlaspietea rotundifolii* phytosociological class. As plant cover increases, species richness also rises, and communities shift toward basophilic grasslands, notably of *Elyno-Seslerietea* class. Soil properties change accordingly, with increases in depth, stability (shear vane strength), organic matter, and nutrient content. These findings suggest that plant cover can serve as a reliable and cost-effective proxy for assessing soil development and vegetation characteristics in alpine scree environments. This approach may support monitoring and restoration strategies aimed at conserving biodiversity and ecosystem function in mountain and alpine landscapes facing increasing environmental pressures.

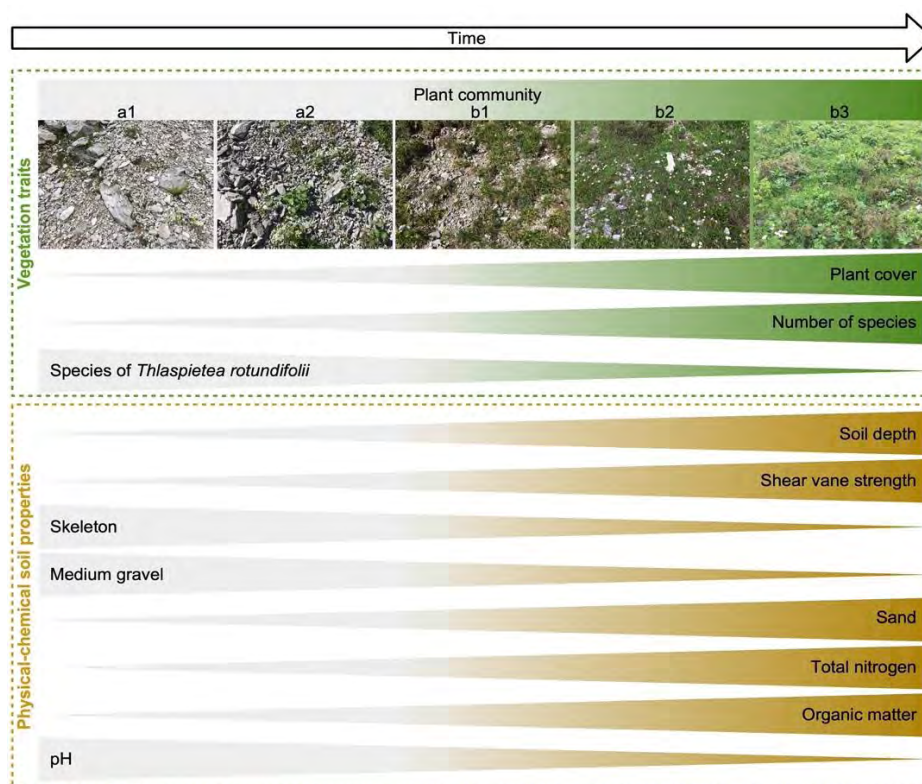


Fig. 1. Impacts of limestone scree colonization on vegetation traits and physical–chemical soil properties considered in this research. For each factor, its change during ecological succession is illustrated schematically

1) L. Giupponi, V. Leoni, D. Pedrali, M. Zuccolo, A. Cislighi (2022) Plant and Soil, 483, 495-513

9.10 = Towards adaptive food systems: germination behaviour and climate resilience of two endemic Mediterranean *Brassica oleracea* wild relatives

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Wild Edible Plants (WEPs) and Crop Wild Relatives (CWRs) represent a vital reservoir of genetic, ecological, and cultural diversity, increasingly recognized as essential for the adaptation and diversification of global food systems in the face of climate change. The Mediterranean Basin is a hotspot for such resources, hosting over 2,700 WEPs, including numerous species in the genus *Brassica*, one of the most species-rich and agriculturally relevant genus.

In this study, we investigated the seed ecology and climate resilience of *Brassica insularis* Moris and *Brassica tyrrhena* Giotto, Piccitto & Arrigoni, two endemic wild relatives of cultivated cabbages (*Brassica oleracea* L.) from Sardinia (Italy), with limited distribution and recognized ethnobotanical value. Seeds were collected from two natural populations per species, and germination responses were tested across eight constant temperatures (5°C to 45°C) to determine base (T_b), optimal (T_o) and ceiling (T_c) temperatures. A thermal time approach was applied to quantify temperature-dependent germination rates and assess physiological thresholds. Additionally, species distribution models were used to project the impact of climate change on their potential range.

Both species displayed high germination performance under thermal stress, suggesting an inherent tolerance to elevated temperatures. By combining ethnobotanical relevance with functional germination traits, our results highlight the dual potential of these species: as candidates for crop improvement or for de novo domestication in a rapidly changing climate. This interdisciplinary approach offers a replicable model for evaluating the adaptive potential of other WEPs and CWRs.

9.10 = Urban reforestation and human health: the role of allergenic species in Rome

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The observable climate changes seem to be more evident in urban environments. The urban heat island effect, in fact, amplifies the extent and impact of these variations: from the point of view of human health, this determines a worsening of living conditions for people, and in particular for children and the elderly (1); from the phenological point of view, through aerobiological monitoring, it is possible to record variations in terms of phenological indicators and production indicators. It is in fact possible, having long series of data available, to evaluate any variations in terms of the start, duration and end of flowering and variations in terms of pollen production (2). One of the possible actions capable of mitigating the increase in temperatures in urban environments is the reforestation of cities. Rome municipality in collaboration with (3) carried out a study to project and manage tree lines of the city according to different purposes. The preliminary study considered only 200 streets but is ongoing. According to this study along the streets of Rome are present about 120,000 trees. Moreover, we should consider all the trees present in parks, gardens and villas. The number of trees in Rome is very high, more than 400,000. Trees in the city has several roles, such as mitigating air pollution (capture of CO₂, reduction of pollutants), reducing the heat island, increasing biodiversity, mitigating noise pollution, improve environments quality. The city of Rome has also started urban reforestation projects. According to PNRR about 1 million trees and shrubs will be planted in Rome, following ecological criteria. The choice of species considers multiple factors, including that of avoiding the introduction of allergenic species. However, if you look at the list of species used or in use, you will notice that there are some very allergenic ones, such as cypress, olive and plane, hazel, hornbeam etc. Examining the data available at the "Tor Vergata" Aerobiological Monitoring Center, collected since 1996, it is possible to note how some common species responsible of arboreal pollen are allergenic causing import diseases in population. For example, pollen belonging to Cupressaceae is about 25% of total pollen in Rome.

There are three aerobiological monitoring stations operating in the municipality of Rome: Tor Vergata (TV) - in the south-east of the city, outside the GRA ring road, S. Pietro (SP) - in the north, inside the GRA ring road, and Cipro (CP) - in the city center. A comparison of the averages of the parameters describing the phenological indicators (start, end, and duration of flowering) and production (APIn and maximum peak) between the three monitoring stations in the city of Rome shows that the TV station presents clear differences (t-Student p value < 0.05) compared to the data recorded for the SP and CP stations. Furthermore, dividing the monitoring years into two periods (1997-2010 and 2011-2024), for the TV and SP monitoring stations (data not available for CP as it has been active since 2018), differences can be seen only in the SP station: from the comparison of the averages of the parameters describing the phenological indicators (start, end, and duration of flowering) and production (APIn and maximum peak), the values of the t-Student coefficient are significant (p<0.05) in the case of the end of the pollen season, duration of the pollen season, APIn, and maximum peak for Cupressaceae and Platanaceae; in particular, for Cupressaceae, it can be seen that although the duration of the pollen season tends to lengthen, the amount of airborne pollen recorded is decreasing.

The choice of unsuitable species could reverse the trend, contributing to a worsening in environmental quality. We hope for a rethink or a limitation in the use of these species.

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9.12 = Seed conservation and germination ecology of *Eokochia saxicola* (Guss.) Freitag & G.Kadereit across its distribution area

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Eokochia saxicola [syn. *Bassia saxicola* (Guss.) A.J. Scott] is an endemic Amaranthaceae species restricted to the South Tyrrhenian region, currently known from only three locations: Capri Island, the promontory of Capo Palinuro, and Strombolicchio islet (Aeolian Islands). It is a chasmophyte, from a recently established monospecific genus described by Kadereit & Freitag (2011), and inhabits north-facing coastal cliffs on both limestone and basaltic substrates. Due to its unfavourable conservation status (“EN”, according to IUCN criteria), *E. saxicola* has been selected as a target species in the LIFE SEEDFORCE project. Its fruits are enclosed in a persistent perianth. The seeds have well-differentiated, green-coloured embryos and can tolerate exposure to sea water, but lose viability within four months after collection (1). This study investigated the effect of several environmental and biological factors on seed germination behaviour: pH, temperature, salinity, osmotic potential, seed maturity and presence or absence of the perianth. Experiments were conducted on seeds from the three known populations to assess whether isolation has led to adaptive divergence in germination niche. Additionally, seed viability was tested after storage at different relative humidity (RH) levels for 1, 3 and 7 months. Substrate pH did not affect germination across the three populations. Temperature, however, had a significant effect: germination was highest between 5 and 15°C, particularly under alternating temperatures regime, and was strongly inhibited above 20°C. Seeds exposed to 150 mg/L NaCl germinated at rates similar to the control across the three populations. An osmotic potential below -0.8 MPa drastically reduced germination, which is recovered as soon as the osmotic stress is relieved. Ripe seeds with brown perianth had higher germination than unripe seeds with green perianth. Removing perianth improved both germination speed and final percentage. After 7 months of storage at 15% and 30% RH, seeds retained the ability to germinate but failed to reach the seedling stage, as cotyledon expansion did not occur. Storage at 50% RH led to a significant loss of viability within just one month. In conclusion, *E. saxicola* exhibits a germination pattern typical of Mediterranean coastal species, preferring low and alternating temperatures and showing limited salinity tolerance. Regarding desiccation tolerance, its seeds display intermediate behaviour and are therefore unsuitable for long-term conservation under standard seed bank conditions (15%RH, -20°C). Our results highlight the importance of coordinated efforts between germplasm banks and botanical gardens for the *ex-situ* propagation and cultivation of *E. saxicola*, as well as the urgent need to strengthen the *in-situ* conservation perspectives of this highly localised Mediterranean endemic.

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9.13 = *Apium repens* (Jacq.) Lag. in Slovenia – from extinction to reintroduction

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Creeping marshwort (*Apium repens* (Jacq.) Lag.) is a perennial creeping plant that forms rosettes of simple pinnate leaves from which stolons or runners form and rooting at the nodes. It is a species listed in Annex II of the Habitats Directive, and it is rare and endangered in many European countries. This is due to its special habitats: River mudflats, gravel banks and flood-prone wet meadows. The last reliable records in Slovenia are more than 150 years old. Exemplars discovered in 1997 are doubtful, so that the species was considered extinct in Slovenia. This was the reason for the reintroduction of the species to its natural habitat on the river Drava. First, the area along the middle course of the Drava near Središče was thoroughly explored, also to identify the most suitable habitats. Since no population of *A. repens* was found, the nearest site (Slunjščica river spring, Croatia) was identified and 20 vegetative parts, which were part of a larger population of at least 200 specimens, and seeds were officially exported to Slovenia.

To obtain sufficient plants for further reintroduction, a standardised tissue culture protocol for micropropagation of *A. repens* by axillary shoots and stolons was developed (Figure 1). Collected genotypes cultured on solid Murashige and Skoog medium (MS) developed the highest number of shoots and stolons when 2 μ M BAP were used. Regenerated shoots rooted on media with and without growth regulators and transferred to the greenhouse and subsequently the natural outdoor conditions.

Reintroduction of 80 plants on polygons with a total size of 0.28 ha was performed at one site between Ptuj and the Zavrč border crossing and at six sites located between Lake Ormož and Središče ob Dravi (Figure 2 and 3). A two-year monitoring revealed that plants were flowering and produced seeds. However, not all exemplars survived, especially after exceptional floods in 2023. Nevertheless, the seed production and high ability of vegetative reproduction are promising for a long-term persistence in the area.



Fig. 1. *Apium repens* in tissue culture



Fig. 2. Reintroduction sites



Fig. 3. Reintroduction polygons (blue) along the River Drava between Lake Ormož and Središče ob Dravi

Free Topic = Effects of sunscreen compound nano-TiO₂ on the intertidal species *Ericaria amentacea* (Fucales, Phaeophyceae)

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The brown macroalga *Ericaria amentacea* (C. Agardh) Molinari & Guiry is a key intertidal habitat-forming species endemic to the Mediterranean Sea, providing critical ecosystem services, including habitat structuring and primary productivity (1). However, like other *Cystoseira* s.l. species, it is undergoing a severe decline due to eutrophication, physical disturbances such as trampling, and exposure to emerging chemical pollutants. Among these, nano-sized titanium dioxide (nano-TiO₂) is a pervasive contaminant derived from personal care products, widely used for its UV-blocking properties. While considered safe for human use, growing evidence indicates its toxicity to marine life (2).

This study assesses the vulnerability of *E. amentacea* to nano-TiO₂ exposure. Both adult specimens and seedlings were exposed to different concentrations of nano-TiO₂ (0, 5, 50, and 500 µg/L) over a three-week period. Effects were evaluated at multiple biological levels, including metabolic and photosynthetic performance, intracellular nanoparticle accumulation, and key biochemical stress markers (chlorophyll-a, phaeophytin, hydrogen peroxide (H₂O₂), superoxide dismutase (SOD), and ascorbic acid).

Our findings indicate a dose-dependent effect in photosynthetic performances and metabolic rates. Within 10 days, photosynthetic efficiency (PSII quantum yield, Fv/Fm) significantly decreased from 0.64 ± 0.03 (control) to 0.58 ± 0.02 (500 µg/L treatment). After three weeks, oxygen production in adult thalli was reduced by half at 500 µg/L compared to the controls. Additionally, oxidative stress markers—including elevated H₂O₂ levels, increased SOD activity, and phaeophytin accumulation—were detected across all tested concentrations, with seedlings exhibiting heightened sensitivity even at the lowest exposure level.

Understanding the long-term ecological consequences of this ubiquitous yet understudied contaminant is crucial for informing conservation strategies aimed at safeguarding Mediterranean coastal biodiversity.

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Free Topic = Effects of urban soil components on germination and early development of *Borago officinalis*: implications for urban green space management

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Urban vegetation is subjected to intense environmental stress and human disturbance, which significantly affect plant growth and development (1). Urban soils are often characterized by altered hydrology, surface sealing, poor quality, and high levels of pollutants (2). Compared to rural or natural ones, urban soils differ significantly in structure, composition, and function: they are typically drier, more compacted, poorly drained, and alkaline (3). Moreover, these soils frequently contain a heterogeneous mixture of natural materials and anthropogenic components, such as construction debris, asphalt residues, and industrial byproducts (e.g., waste foundry sand), which further modify soil texture, porosity, and nutrient availability (3). Although interest in the effects of urban soil substrates, such as asphalt and foundry sand, on plant germination and growth is growing, research in this field remains limited and current studies emphasize the need for targeted experiments to clarify how specific soil components influence seed germination and seedling development in urban environments. This study aimed to experimentally assess the impact of typical urban soil components (asphalt and foundry sand) on the germination and early development of *Borago officinalis*. The experiment was performed in greenhouse with no environmental control. Seeds were sown in polystyrene trays filled with four substrates commonly found in cities: a) asphalt, b) standard foundry sand, c) recycled foundry sand, and d) commercial potting soil. Germination success, seedling growth, and mortality were monitored to generate growth patterns for each substrate. Furthermore, plant morpho-anatomical and physiological traits, including leaf area, fresh and dry biomass of roots and shoots, stomatal density, stomatal conductance and photochemical efficiency, were measured and compared across treatments to provide a comprehensive evaluation of substrate effects on plant development. We found that, although germination rates were high across all treatments (above 70%), only a few seedlings grown on asphalt developed both cotyledons and leaves, while a significant higher proportion of seedlings grown in potting soil reached these developmental stages. Mortality rates were significantly higher in the asphalt treatment compared to the others, and growth patterns indicated slower leaf development in asphalt than in soil. Plants grown in soil also exhibited significantly greater biomass (both root and aerial parts) compared to those grown in standard foundry sand and asphalt. Additionally, stomatal conductance and transpiration were highest in the leaves of plants grown in soil, which also had lowest stomatal density. Interestingly plants grown on asphalt and in standard foundry sand showed highest stomatal conductance and transpiration rates in cotyledons. Overall, results highlighted how specific urban soil component can influence plant establishment and development, an aspect that should be considered in the management of the urban green spaces. Our findings align with the aims of Spoke 5 of the National Biodiversity Future Centre, which “focuses on urban biodiversity and seeks solutions to enhance quality of life in cities through the conservation and sustainable management of urban ecosystems” and contribute to a deeper understanding of plant responses to urbanization, ultimately providing valuable insights for a sustainable management of urban biodiversity.

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Free Topic = Insights on the systematics of a peculiar *Armeria* (Plumbaginaceae) population Sardinia

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The genus *Armeria* Willd. (Plumbaginaceae) encompasses approximately 108 species of perennial, diploid herbs, with its primary centre of diversity located in the Mediterranean Basin (1). The taxonomic delimitation within *Armeria* is notably challenging due to several interrelated factors: a high degree of intraspecific morphological variability, a relatively uniform vegetative architecture (typically characterized by basal rosettes of linear-lanceolate leaves and scapose flowering stems terminating in a solitary capitulum), and the presence of weak reproductive barriers between co-occurring taxa (2). A recent integrative taxonomic study of *Armeria* in the Cyrno-Sardinian biogeographic province (2) led to the recognition of five distinct species: three narrowly endemic to Sardinia (*A. sulcitana* Arrigoni, *A. sardoa* Spreng., and *A. morisii* Boiss.) and two endemic to Corsica (*A. leucocephala* Salzm. ex W.D.J.Koch and *A. soleirolii* (Duby) Godr.). During a floristic survey in the Ogliastro region (Montarbu di Seui, eastern Sardinia), we documented a rupicolous and orophilous population of *Armeria* growing on exposed Mesozoic limestone outcrops. Although this population bears a superficial resemblance to *A. sulcitana* and *A. sardoa*, it exhibits an unusual habit and some potential morphological differences, rendering its identification uncertain.

To clarify the systematics of this population, morphometric data from 12 individuals are being analyzed using the Gaussian Mixture Models framework (3) and compared with the data presented in (2). In addition, karyological and DNA sequence data (ITS and chloroplast regions: *trnF-trnL*, *trnH-psbA*, *trnL-rpl32*, *trnQ-rps16*) are being generated and compared with those published in (2).

Pending the results of a more comprehensive integrative taxonomic assessment, we wish to highlight the pivotal role of targeted floristic exploration in revealing previously unrecognized biodiversity. Such findings are crucial not only for advancing our understanding of species boundaries and evolutionary processes in *Armeria*, but also for shedding light on broader patterns of speciation and biogeographic structuring within Tyrrhenian island floras.

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