

# XVII OPTIMA Meeting

20-23 September 2023 Erice, Italy

# ABSTRACTS







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# ABSTRACTS

Oral Presentations, Posters

*Organized by:* OPTIMA (Organization for the Phyto-Taxonomic Investigation of the Mediterranean Area) University of Palermo *PLANTA* – Mediterranean Center for Research, Documentation and Training

### OPTIMA (Organization for the Phyto-Taxonomic Investigation of the Mediterranean Area)

### XVII OPTIMA Meeting

20-23 September 2023, Erice, Italy

### Abstracts

Oral Presentations, Posters

Editors: Gianniantonio Domina Francesco M. Raimondo, Werner Greuter Technical editing: Gianniantonio Domina

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#### Programme

#### Tuesday 19 September [Palermo, Via Archirafi 38]

10h00–18h00 – Meetings of the Scientific commissions of OPTIMA – Section of Section of Botany, Zoology and Anthropology of the Department STEBICEF.

18h15 - Bus transfer to Erice.

#### Wednesday 20 September [Erice]

9h00 - Welcoming of the participants and registration - Instituto San Rocco.

- 9h00 Joint Meeting of the Executive Council & International Board of OPTIMA (restricted, *Instituto San Rocco*).
- 11h00 Opening Ceremony Auditorium Pam Dirac.
- 12h00 **Plenary conference** *Auditorium Pam Dirac* K. H. C. Başer (Nicosia, Cyprus) : Essential oils: science, technology, and applications, yesterday and today.
- 13h00 Lunch Palazzo Sales.
- 14h30 *Symposium* 1 *Palazzo Sales* Mediterranean refugia: importance for plant conservation.
- Chairs: F. Médail (Marseille, France), M. Fennane (Rabat, Morocco)
- 14h30–15h00. <u>A. Baumel</u>, F. Médail, G. Nieto Feliner, S. La Malfa, M. Di Guardo, M. Bou-Dagher Kharrat, L. Ouahmane, K. Diadema, H. Sanguin, J. Viruel: Late Pleistocene expansions from a single climatic refugium and close wild–domesticated relationships in the carob tree.
- 15h00–15h30. M. Guerrina, D. Dagnino, L. Minuto, F. Médail, <u>G. Casazza</u>: Local refugia and environmental heterogeneity: a case study in a Mediterranean mountain system (Southern Alps).
- 15h30–16h00. <u>T. Villaverde</u>, M. Rincón-Barrado, M. Mairal, V. Culshaw, S. Olsson, L. Pokorny, R. Riina, I. Sanmartín: The Rand Flora pattern: from micro to macroevolutionary levels.
- 16h00–16h30. <u>M. Fois</u>, E. Farris, G. Bacchetta: The floristic diversity of Sardinia: a starting point for the implementation of phylogeographic, ecological and conservation studies in the Mediterranean Basin.

16h30-17h00. Coffee Break

- 17h30–18h00. <u>I. Rešetnik</u>, S. Španiel, M. Temunović, S. Bogdanović, I. Ljubičić, A. Terlević, M. Mucko: Multiple Balkan glacial refugia - an overview of genetic imprints.
- 18h00–18h15. E. Di Gristina, E. Bajona, <u>G. Domina</u>, F. M. Raimondo : The Trapani mountains (NW Sicily), refuge for relict or highly localized endemic plants.
- 18h15–18h30. <u>F. Bartoli</u>, G. Zangari, M. Iocchi, F. Lucchese : Latium Flora: Distribution model of species with conservationist interest in refugia.
- 18h30–18h45. I. Camarda: Intérêt phytogénétique et endémismes du massif du Gennargentu (Sardaigne).
- 14h30 *Symposium 2 Instituto San Rocco* Taxonomy, ecology and conservation of Mediterranean fungi, lichens and bryophytes.
- Chairs: M. Ros (Murcia, Spain), G. Venturella (Palermo, Italy), P. Campisi (Palermo, Italy), S. Ravera (Palermo, Italy)
- 14h30–15h00. <u>A. De Agostini</u>, M. Aleffi, A. Cogoni, S. Poponessi : Bryophytes in small circum-Sardinian islands: biogeographic patterns, ecology and comparison with other small circum-Italian islands.
- 15h00–15h30. J. G. Segarra-Moragues : Progress in the taxonomical and evolutionary knowledge of the aquatic liverworts *Riella*.
- 15h30–16h00. <u>N. Hodgetts</u>, C. Views: Mediterranean bryophytes: are there any effective conservation strategies?
- 16h00–16h30. N. G. Medina: Decoding the determinants of the abundance and distribution of Mediterranean bryophytes.

16h30-17h00. Coffee Break

- 17h00–17h30. <u>A. Guttová</u>, M. Svitok, Z. Fačkovcová, L. Paoli, J. K., M. Slovák, S. Munzi, J. Breidy, H. Dokmak, D. Senko : In search of strictly Mediterranean lichens: contribution of collection based approach to characterization of European Mediterranean.
- 17h30–18h00. <u>P. Campisi</u>, M. Puglisi: Threatened Liverworts of Italian bryoflora: analisys of biological and ecological characters.
- 18h00–18h30. G. Mirabile: Biodiversity of fungi in freshwater ecosystems of Italy.

#### 20h00. Welcome cocktail.

#### Thursday 21 September [Erice]

9h00 - Symposium 3 - Palazzo Sales - Biosystematics in the Mediterranean area 1.

- Chairs: K. Marhold (Bratislava, Slovak Republic), C. Oberprieler (Regensburg, Germany), L. Peruzzi (Pisa, Italy), P. Bareka (Athens, Greece)
- 9h00–9h20. M. Šlenker, A. Kantor, J. Zozomová-Lihová, J. Kučera, K. Skokanová, B. Šingliarová, T. Mandáková, <u>K. Marhold</u>: Balkan diploids and polyploids in *Cardamine* (*Brassicaceae*).
- 9h20–9h40. <u>I. Álvarez</u>, J. Fuertes-Aguilar, S. Garcia, T. Garnatje, O. Hidalgo, G. Nieto Feliner, J. Pellicer, M. Rosato, J. A. Rosselló, R. Torices, J. Vallès, D. Vitales: The genus *Anacyclus* (*Anthemideae*, *Asteraceae*) as a model for research on diploid hybrid lineages evolution.
- 9h40–10h00. <u>M. Tiburtini</u>, L. Peruzzi: Morphometric tools to test species delimitation: the case of *Armeria (Plumbaginaceae)* in central Mediterranean area.

- 10h00–10h20. D. Caković, M. Boschin, E. Záveská, M. Skubic, R. El Mokni, <u>B. Frajman</u>: Disentangling relationships in different Mediterranean *Euphorbia* lineages (*Euphorbiaceae*) using integrative approach.
- 10h20–10h40. D. Harpke: Towards a resolved phylogeny for a new classification of *Crocus* (*Iridaceae*).
- 10h40–11h00. <u>M. Dorfner</u>, C. Oberprieler: From error-prone Nanopore reads to valuable insights: AFLP-based genome reduction and the SLANG pipeline for multi-locus analysis in biosystematic research.
- 11h00–11h30. Coffee Break
- 11h30–11h50. <u>M. Guerrina</u>, L. Varaldo, A. Baumel, L. Minuto, A. Giacò, L. Peruzzi, L. Sáez, R. Carballal, P. Caputo, D. De Luca, G. Bacchetta, L. Podda, F. Conti, F. Bartolucci, G. Casazza : Exploring the phylogenetic relationships in *Santolina (Asteraceae)*: a taxonomically complex genus endemic to the Mediterranean Basin.
- 11h50–12h10. <u>D. Gutiérrez-Larruscain</u>, P. Vargas, M. Fernández-Mazuecos, J. G. Pausas: The evolutionary history of junipers (*Juniperus* sect. *Juniperus*) based on genomic (GBS) analysis.
- 12h10–12h30. <u>J. Franzoni</u>, G. Bacchetta, F. Conti, G. Domina, S. Fior, L. Minuto, L. Peruzzi: Tackling taxonomic inflation: biosystematics studies of the *Dianthus virgineus* complex (*Caryophyllaceae*) in the Central Mediterranean
- 12h30–12h50. S. Casavecchia, M. Giovannotti, F. Carducci, M. A. Biscotti, M. Barucca, E. Trucchi, F. Giannelli, G. Quattrini, S. Pesaresi, L. Aquilanti: Diversity of the Italian populations of *Onopordum tauricum (Asteraceae)* as revealed by ddRAD sequencing and morphometric data analysis.
- 9h00 *Symposium* 4 *Instituto San Rocco* Taxonomy, ecology and conservation of Mediterranean marine and freshwater algae.
- Chairs: A. M. Mannino (Palermo, Italy), C. Rodriguez-Prieto (Girona, Spain), M. Cantonati (Bologna, Italy)
- 9h00–9h30. A. Falace: The challenge of conservation and restoration of macroalgal forests with special attention to the Mediterranean.
- 9h30–10h00. <u>C. Rodríguez-Prieto</u>, J. Morcillo: What's new in *Acrosymphytales* (*Rhodophyta*)? The case of Mediterranean *Acrosymphyton purpuriferum*.
- 10h00–10h30. <u>K. M. Manoylov</u>, R. Stancheva, M. Cantonati: Understanding the invisible population and community ecology of algae from Mediterranean streams on two continents.
- 10h30–11h00. <u>M. Cid Rodriguez</u>, O. Bilous, G. Dörflinger, H. Lange-Bertalot, A. Papatheodoulou, A. A. Saber, D. Spitale, M. Cantonati: New taxonomic and ecological data on new-to-science, rare and insufficiently known *Nitzschia* species (*Bacillariophyta*) from the Island of Cyprus.
- 11h00–11h30. Coffee Break
- 11h30–12h00. M. Cantonati: Ecology, conservation, and taxonomy of diatoms in Mediterranean streams and springs in the face of climate change.

- 12h00–12h30. F. Rindi: Biodiversity associated with Mediterranean fucalean forests and mechanisms regulating it: an assessment of the current knowledge.
- 12h30–13h00. M. Morabito: DNA barcodes as permanent labelling in floristic and systematic studies of macroalgae.
- 13h15 Lunch Palazzo Sales.
- 14h30 Symposium 5 Palazzo Sales Biosystematics in the Mediterranean area 2.
- Chairs: K. Marhold (Bratislava, Slovak Republic), C. Oberprieler (Regensburg, Germany), L. Peruzzi (Pisa, Italy), P. Bareka (Athens, Greece)
- 14h30–14h50. <u>M. Escudero</u>, A. Valdés-Florido, E. Maguilla, V. Simón-Porcar, J. Arroyo: Patterns and mechanisms of plant diversification driven by chromosome shifts in Mediterranean lineages.
- 14h50–15h10. <u>K. Koutroumpa</u>, E. Conti, N. Kilian: Taxonomic and evolutionary studies on Mediterranean plant groups in Greece: the examples of *Centaurea (Asteraceae)* and *Limonium (Plumbaginaceae)*.
- 15h10–15h30. I. Rúrik, A. Melichárková, J. Kučera, E. Gbúrová Štubňová, J. Kochjarová, O. Paun, P. Vdačný, <u>M. Slovák</u>: Drivers of diversification, speciation, and trait evolution in the European Alpine System endemic genus *Soldanella (Primulaceae)*.
- 15h30–15h50. <u>S. Tomasello</u>, E. Manzo: Phylogenomics and the biogeography of the Old-World *Xanthium strumarium (Asteraceae)*.
- 16h00-16h30. Coffee Break
- 16h30–16h50. <u>A. Giacò</u>, L. Peruzzi: Lumping and splitting: how an integrative approach can guide taxonomists in species delimitation.
- 16h50–17h10. <u>A. Maccioni</u>, S. Macis, M. Gibernau, E. Farris: Intra- and inter-population phytochemical variability of *Teucrium marum (Lamiaceae)* in Sardinia: relevant for chemotaxonomy or just determined by ecological factors?
- 17h10–17h30. <u>C. Chatellier</u>, C. Tollon, B. Khadari, J. Molina: Towards defining the genetic statute of *Limonium cuspidatum (Plumbaginaceae)*, a protected taxon in South French, using microsatellite markers.
- 14h30 *Symposium* 6 *Instituto San Rocco* Ecosystems restoration and species conservation.
- Chairs: J. M. Iriondo (Madrid, Spain), P. Marino (Palermo, Italy)
- 14h30–15h00. P. Fraga i Arguimbau : More than 20 years of flora conservation in Menorca: landscape changes and management.
- 15h00–15h30. <u>A. García-Fernández</u> S. Sacristán-Bajo, E. Torres, C. Lara-Romero, M. Luisa Rubio Teso, J. M. Iriondo: Facilitated adaptation as a conservation tool for plant adaptation to climate change
- 15h30–16h00. <u>M. L. Rubio Teso</u>, G. Asens, A. García-Fernández, C. Lara-Romero, A. Molina, C. del Tío Navas, E. Torres, J. M. Iriondo : Design and development of genetic reserves for plant conservation.

16h00-16h30. Coffee Break

- 16h30–17h00. H. Leschner : Celebrating a centennial 100 years of struggle for the preservation and rehabilitation of plants in Israel.
- 17h00–17h15. V. Gianguzzi, <u>G. Barone</u>, E. Di Gristina, F. Sottile, G. Domina: Use of micropropagation techniques for the conservation of endangered species with reproductive problems.
- 17h15–17h30. <u>A. Prigioniero</u>, M. Tartaglia, D. Zuzolo, M. A. Ranauda, P. Scarano, C. Guarino : Populations genomic analysis of *Primula palinuri (Primulaceae)*, a narrow endemic Mediterranean plant species on the edge of precipice.
- 17h30 *Roundtable Palazzo Sales* World Flora Online, Euro+Med, APD, inaturalist, efloraMaghreb, how to get ahead, how to structure the synergies with databases.
- Chair: C. Chatelain (Genéve, Switzerland)

#### Friday 22 September [Erice]

- 9h00 *Symposium* 7 *Instituto San Rocco* Herbaria and the understanding of Mediterranean plant diversity: past, present and future.
- Chairs: M. Carine (London, U.K.), G. Domina (Palermo, Italy)
- 9h00-9h30. G. Cristofolini: Origin and Evolution of Herbaria in the 16th Century.
- 9h30–10h00. <u>A. Stefanaki</u>, T. van Andel: How to approach the unknown? Elucidating the origins of the 16th-century En Tibi and Rauwolf book herbaria.
- 10h00–10h30. P. Muñoz-Rodríguez: Taxonomy in the spotlight: integrating herbarium-based research and genomics to monograph the world's plants.

10h30-11h00. Coffee Break

- 11h00–11h30. <u>C. Chatelain</u>, F. Mombrial: Herbaria for the next generations, the exemple of Litardière herbarium and herbonautes in M.
- 11h30–12h00. <u>F. Willems</u>, J. F. Scheepens, O. Bossdorf: Forest wildflowers bloom earlier as Europe warms: lessons from herbaria and spatial modelling.
- 12h00–12h30. F. Roma-Marzio, D. Dolci, G. Astuti, N. Magrini, F. Pierotti, S. Maccioni, R. Vangelisti, L. Amadei, <u>L. Peruzzi</u>: Digitization of historical herbaria. A case study from the Herbarium of Michele Guadagno at Pisa (PI).
- 12h30–13h00. W. Addink: Herbarium specimens in the web of scientific data in 2030, a forward look.
- 8h30 Symposium 8 Palazzo Sales Useful plants in the Mediterranean Countries.
- Chairs: K. H. C. Başer (Nicosia, Cyprus), G. A. Malfa (Catania, Italy), E. Kožuharova (Sofia, Bulgaria)
- 8h30–9h00. <u>N. Smyth</u>, J. Stout, K. Chandler, Ú. Fitz Patrick : Building pollinator resources in urban areas pollinator friendly plants for different situations.
- 9h00–9h15. P. Zhelev: Diversity and importance of arboreal plants.
- 9h15–9h30. F. Palla: Film Plants of the Lamiaceae family as source of bioactive derivatives.
- 9h30-9h45. A. Dafni, B. Boeck : Medicinal Plants of the Bible Past, Present, and Future.
- 9h45–10h00. <u>G. Benítez</u>, L. Aceituno-Mata, M. Pardo-de-Santayana : A review of the dye plants in Spain.
- 10h00–10h15. I. Aneva, Wild plants used as spices.

- 10h15–10h30. A. R. M. S. Al Tawaha: Edible Treasures: Discovering the Riches of Wild Food Plants.
- 10h30-11h00. Coffee Break
- 11h00 Symposium 9 Palazzo Sales Health Benefits of Wild Mediterranean Plants.
- Chairs: K. H. C. Başer (Nicosia, Cyprus), R. Acquaviva (Catania, Italy), V. Spadaro (Palermo, Italy)
- 11h00-11h20. M. Koşar, K. H. C. Başer: Saffron (Crocus sativus, Iridaceae) in ocular diseases.
- 11h20–11h40. <u>P. Malaspina</u>, F. Polito, L. Cornara, V. De Feo, V. Spadaro, N. Miceli, F. M. Raimondo: Pharmacognostic characterization of *Diplotaxis tenuifolia (Brassicaceae)* from Campania and Sicily (Southern Italy).
- 11h40–12h00. D. Orčić, S. Berežni, <u>N. Mimica-Dukić</u>: *Anthriscus sylvestris (Apiaceae)*: Phytochemical and pharmacological approach.
- 12h00–12h15. <u>G. A. Malfa</u>, S. Bianchi, C. Di Giacomo, B. Tomasello, P. Marino, R. Acquaviva: Phytochemical constituents, biological activities, and health-promoting effects of Sicilian wild artichokes.
- 12h15–12h30. <u>E. Bedir</u>, M. Küçüksolak, S. Yılmaz, G. Üner, P. Ballar Kırmızıbayrak: Utilization of endophytic fungi for biotransformation to obtain bioactive compounds.
- 12h30–12h45. L. Giamperi, P. Marino, A. E. A. Bucchini: Enhancement of ancient fruit varieties registered in the regional repertoires through the study of their health properties.
- 12h45–13h00. <u>A. E. A. Bucchini</u>, F. Palla, L. Giamperi : Antioxidant and anti-inflammatory activity of ancient apple varieties from central Italy.

13h00 – Lunch – Palazzo Sales.

- 14h30 Poster Session-Instituto San Rocco.
- 16h30-16h30. Coffee Break
- 16h30 *Roundtable Auditorium Pam Dirac* Plant diversity and restoration of the historic garden «Giardino del Balio» (Erice) and presentation of the project for its restoration [financed by the Italian Ministry of Culture with funds from the PNRR].

Chair: J. Wade (Cernobbio, Italy)

#### 20h00 – Congress Dinner

#### Saturday 23 September [Erice]

- 9h00 *Symposium* 10 *Instituto San Rocco* Crop Wild Relatives in the Mediterranean countries (Symposium dedicated to the memory of Professor V. H. Heywood).
- Chairs: B. Valdès (Sevilla, Spain), A. Giovino (Palermo, Italy), M. Rejdali (Temara, Morocco) 9h00–9h30. Introduction. To the memory of Professor Vernon Hilton Heywood.
- 9h30–10h00. B. Valdés: Recent introduction into cultivation of wild plants in the Iberian Peninsula.
- 10h00–10h30. A. Amri, S. K. Agrawal, F. Bassi, M. Sanchez-Garcia, M. El-Bouhssini, <u>M. Yazbek</u>, H. Aberkane: Conservation and use of wild relatives of cereals and food legumes.

#### 10h30-11h00. Coffee Break

- 11h00–11h30. E. Kozuharova, I. Aneva: Introduction into cultivation of the Balkan endemic *Sideritis scardica (Lamiaceae).*
- 11h30–12h00. O. Barazani: Applying genomic tools in the study of Crop Wild Relatives in the south Levant.
- 12h00–12h30. <u>F. Bonanno</u>, A. Giovino, A. Marchese, S. Aprile, F. M. Raimondo: Taxonomic and genetic diversity of the Sicilian relatives of azarole (*Crataegus azarolus, Rosaceae*).
- 12h30–13h00. <u>P. Marino</u>, V. Spadaro & G. Zizzo: Taxonomy, ecology and distribution of the Sicilian relatives of artichoke (*Cynara scolymus*, *Asteraceae*).
- 9h00 *Symposium* 11 *Palazzo Sales* Invasive plants and their impact in Mediterranean countries.
- Chairs: O. Vasic (Beograd, Serbia), F.M. Raimondo (Palermo, Italy)
- 9h00–9h30. C. Gómez-Bellver, N. Ibáñez, N. Nualart, J. López-Pujol: Cataloguing the alien flora of north-western Mediterranean basin: recent efforts and future prospects.
- 9h30–10h00. <u>V. Lozano</u>, G. Brundu: Invasive alien plants of European Union concern in Italy: distribution and threats.
- 10h00-10h30. M. Rat: Changing perceptions of invasive plants in Serbia.

#### 10h30-11h00. Coffee Break

- 11h00–11h30. A. M. Mannino: An overview on alien macrophytes in Sicilian Marine Protected Areas (Southern Mediterranean Sea).
- 11h30–12h00 <u>C. Gómez-Bellver</u>, R. Melero, Jordi López-Pujol, N. Ibáñez, N. Nualart, R. Rodríguez-González, A. Bosch-Guiu, D. Vitales, M. Guirado, P. Feliu, G. Carrión, F. Caralt, J. M. Pagès, S. Garcia: LIFE medCLIFFS: towards an integrative management of invasive plant species in the Costa Brava (North-east Iberian Peninsula).
- 12h00–12h30. P. Fraga i Arguimbau: Invasive alien plants in Menorca (Balearic Islands, Spain): characterization, management experiences and proposals for a sustainable control.
- 12h30–12h45. M. L. Gargano, <u>R. Pardi</u>, V. Malacrinò: Invasive trend of *Parkinsonia aculeata (Fabaceae)* in Mediterranean Italy.
- 12h45–13h00. N. Lachashvili, <u>K. Kereselidze</u>, G. Nakhutsrishvili: *Koelreuteria paniculata (Sapindaceae)*, casual alien tree in the South Caucasus and in the Mediterranean region.
- 13h00 Lunch Palazzo Sales.
- 15h00 *Palazzo Sales* Special Event UNESCO site and geopark. Aspromonte National Park (Calabria, S-Italy). Were stones meet ancient forest. The case of Valle Infernale, the ancient beech forest.

16h00-16h30. Coffee Break

16h30 - Palazzo Sales - Closing Ceremony and General meeting of OPTIMA.

20h00 - Farewell Cocktail.

### Sunday 24 September [from Erice]

- 8h00 **Post-Meeting excursion** [Natural Reserve Isole dello Stagnone di Marsala: Mothia and Isola Grande].
- 17h00 Bus transfer to Palermo.

**Oral Presentations** 

### Essential oils: technology and applications, yesterday and today

#### K. Hüsnü Can Başer

Near East University, Faculty of Pharmacy, Department of Pharmacognosy, Nicosia, Cyprus. <u>https://www.khcbaser.com;</u> Email: <u>khcbaser@gmail.com</u>

Essential oils are natural volatiles in liquid and semi-solid state found mainly in plants or other organisms. They can be freed from their matrices by a thermal process or expression, and are obtained by water, steam, water and steam distillation, dry distillation or expression only in the case of citrus fruits.

Aromatic chemicals can be captured from the headspace of plant parts emitting volatiles by headspace trapping techniques. Aromatic materials can also be recovered by extracting with organic solvents or fluidized gasses but the resulting material is not technically considered an essential oil but an aromatic extract.

Essential oils are composed of volatile hydrocarbons and their derivatives which may contain O, N, S in their formulae. Essential oils, their fractions and isolated aromachemicals are valuable ingredients of flavour and fragrance, food, perfumery, cosmetics and toiletries, fine chemicals and pharmaceutical industries, and are utilized as such or in diluted forms in therapy or by the aromatherapy sector.

The lecture will highlight the technologies for their production, their utilization and biological activities.

# Late Pleistocene expansions from a single climatic refugium and close wild-domesticated relationships in the carob tree

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Unravelling the origin of present-day populations of plants in relation to the history of Quaternary environmental changes is challenging. In the case of fruit trees, the recurrent exchanges between domesticated and wild populations further complicate reconstructing their phylogeographic history. The carob tree (Ceratonia siliqua L.) is a Mediterranean fruit tree that has played a crucial role in human societies due to its edible fruits, used as fodder for livestock and in subsistence agriculture. The spread of its cultivation and domestication was linked to the development of grafting methods ca. 3,000 years ago. As for several crops, the Middle East and the Eastern Mediterranean regions were initially proposed as the centre of carob domestication. Based on sound biogeographical assumptions and various molecular data and phylogeographic methods, our results refute this hypothesis. After a strong decline the carob tree persisted in a single refugium situated in South Morocco near the Atlantic coast. Phylogenetic divergence supports that range expansion from this refugium occurred before the Last Glacial and along at least two routes. Genetic admixture shows, on the one hand, that domestication took place throughout the Mediterranean region from local populations. On the other hand, there is evidence of genetic influence from the East, probably due to long-distance dispersal of domesticated varieties. These close wild-domesticated relationships over centuries, highlights the importance of wild and semi-natural habitats in climatic refugia for the conservation of fruit tree genetic resources.

# Local refugia and environmental heterogeneity: a case study in a Mediterranean mountain system (Southern Alps)

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Endemic species are of pivotal importance in biogeography and conservation biology, because they are exclusively distributed in a given area. The regions where several endemic species occur are called centres of endemism and are expected to be characterised by stable and often singular climatic conditions that have enable divergence to accumulate over time. In particular, four non-mutually exclusive hypotheses have been proposed to explain accumulation of endemics in the centres of endemism. In general, areas rich in endemics are expected to have climates that are unusual in the region, to have low intra-annual climatic conditions or to have been climatically stable over time. However, within these regions, endemics accumulate in given areas that consequently have an exceptional concentration of endemic species. Several studies aimed at identifying areas rich in endemism, but relatively few studies have attempted to analyse the environmental determinants behind this richness. Here, we aim at identifying the determinants of endemism richness within a centre of endemism, the Southwestern European Alps.

We used distributional data for the endemic plants of SW Alps to explore the patterns of endemism richness to assess how environmental factors contribute to explain these patterns.

Our results suggest that near all hypotheses adequately account for patterns of plant endemics richness in SW Alps. In fact, most of the relationships are statistically significant (Table 2) and the explained variance ranges from weak to substantial. In particular, temperature seasonality, velocity of climate change and the standard deviation of slope have the highest effect.

Overall, no macro-ecological hypothesis fully accounts for species richness, suggesting that, within endemism centres, factors related to species dispersal ability (e.g., the rate of climate change and the standard deviation of slope) as well as region-specific historical factors combine to influence the distributional pattern of endemism richness.

### The Rand Flora pattern: from micro to macroevolutionary levels

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The Rand Flora is a biogeographic pattern that involves various plant lineages exhibiting a disjunct distribution. It is characterized by the occurrence of sister taxa on opposite sides of the African continent, including the coasts of Morocco and Western Sahara, the Horn of Africa, the Eastern Arc Mountains, and southern temperate Africa. This pattern also extends to adjacent areas and archipelagos or islands such as Macaronesia, Socotra, southern Arabia, and Madagascar.

In recent years, several studies have assessed the different mechanisms behind the Rand Flora pattern. Most lineages exhibiting this pattern would have evolved over several million years as a result of a series of aridification events since the Miocene and throughout the Pleistocene. The main processes at work would have been ecological vicariance coupled with climatic extinction, that is, lineages would have been more widespread in the past, not necessarily across the entire distribution range, and underwent fragmentation of their original ranges by the formation of ecological barriers. However, these aridification events were interspersed with wetter periods, secondarily allowing different Rand Flora lineages to expand their ranges by direct long-distance or stepping-stone dispersal. Thus, the Rand Flora pattern was probably formed by a combination of climate-drive extinction, vicariance, and dispersal events that shaped each disjunct distribution differently.

Three Rand Flora examples will be presented: one at the species and population level (*Euphorbia balsamifera*); one at the genus level (*Plocama*); and one at the tribe level (*Camptolomeae*). For *E. balsamifera*, our results show African continental populations are the product of recent migrations (recolonizations) from the Canary Islands (Lanzarote and/or Fuerteventura) to the mainland. Within the genus *Plocama*, divergence times for the Rand Flora clade indicate species from northwest Africa split from those found in southern Arabia ~7 Ma ago, coinciding with the appearance of the Sahara Desert. Since a high conservation of the climatic niche is observed in this Rand Flora clade, ecological niche analyses provide additional support for the ecological vicariance hypothesis as the origin of this disjunction. When considering the tribal level, the current disjunct distribution of *Camptolomeae* across Africa was likely the result of fragmentation, extinction and/or population bottleneck events associated with historical cycles of aridification during the Neogene. The observed pattern of species divergence, progressing from south to north, aligns with the climatic refugia hypothesis. Finally, the Rand Flora disjunction exemplifies biogeographic pseudo-congruence, where multiple asynchronous mechanisms and processes result in the same pattern of distribution.

### The floristic diversity of Sardinia: a starting point for the implementation of phylogeographic, ecological and conservation studies in the Mediterranean Basin

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Sardinia and its ca. 400 satellite minor islands lie in the centre of the Mediterranean Sea, surrounded by the European and African continents, between the Iberian and Italian peninsulas. With a total surface of 24,090 km<sup>2</sup>, it is the largest Mediterranean 'true island' (i.e. an island that was separated from the mainland for at least 20,000 years). Besides its disjunction from the Protoligurian massif around 20 M.a., Sardinian phytodiversity was mainly shaped by the Messinian salinity crisis, Pleistocene climatic and eustatic changes and its high environmental heterogeneity.

We present our studies on the Sardinian floristic diversity. The first focus was on the endemic component, then extended to all taxa of phytogeographic and conservation interest. Of the about 2,300 vascular plant species and subspecies known for Sardinia, we updated the list to include 341 endemic taxa, equal to 15% of the total native flora. Most of them are restricted to Sardinia (57% of endemics) or Sardinia and Corsica (24%), while the rest share their distribution with other islands, mainly from Tuscan, Sicilian and Balearic regions, or with other limited paleogeographically related continental territories. Based on approximately 60,000 georeferenced data, six biogeographical sectors were defined in relation to the diverse local geology and geomorphology. Many taxa are concentrated on mountains, but another large number is in scarcely accessible coastal areas, such as scree and capes and uninhabited minor islands. These areas altogether constitute a network of micro- and nanohotspots of phytodiversity, within the Sardinian meso-hotspot. This information was used to identify climatic and anthropogenic factors as main drivers of current plant diversity. Moreover, it can be used to plan an integrated network of microreserves for plant conservation. However, we depicted several knowledge gaps. For instance, around half of endemics are unassessed according to the IUCN criteria, and this percentage dramatically increases when considering the entire native flora. As regards systematic issues, about 30% of endemics were *inquirenda*, i.e. taxa of taxonomically doubtful validity and/or whose presence in Sardinia requires further investigations. Even less is known about those taxa which are widely but fragmentally distributed and of high regional interest, as in the case of Linaria cossonii Barratte, Gentiana lutea L. or Utricularia vulgaris L., among many others.

This contribution presents the knowledge achievements and gaps in Sardinia, highlighting the need to improve inter- and intra-regional collaborations for more conscious and incisive efforts to ensure such a high plant diversity for next generations.

### Multiple Balkan glacial refugia - an overview of genetic imprints

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The Balkan Peninsula is recognized as an important refugial area for species survival and diversification during Pleistocene climatic cycles. Phylogeographic studies highlight that the topographic complexity and geographic mosaic of habitats favoured the occurrence of multiple disjunct refugia and emphasize the important role these refugia played in shaping the evolutionary history of species. This presentation summarises available phylogenetic and phylogeographic studies of plants from the Balkans that have revealed complex patterns in the geographic distribution of genetic diversity and challenged traditional taxonomic concepts. Within the Balkan Peninsula, a number of taxa show remarkable patterns of phylogeographic concordance involving deep genetic subdivisions, high haplotype richness and distinct hybrid zones. Not only has the Balkan Peninsula sourced the northern redistribution of species after ice ages, but it has also facilitated diversification through patterns of repeated population fragmentation, contraction, expansion and admixture.

Studies in the Balkan Peninsula also indicate the difference in range shifts among taxa with different environmental requirements. Therefore, in the second part of the presentation the phylogeographic pathways of three ecologically divergent and distributionally diverse Balkan plant groups will be outlined. Aurinia saxatilis (Brassicaceae) is a widespread species that ranges from the central, eastern and southern Balkan Peninsula and the adjacent shores of western Asia Minor and the southern Apennine Peninsula, through the Carpathian Mountains to central Europe, growing on rocky grounds and dry soils at low to medium elevations. Dianthus sylvestris (Caryophyllaceae) is distributed across north-western to southern Balkan Peninsula and has a wide elevational range from sea level to high alpine meadows. Festuca bosniaca (Poaceae) is an Apennine-Balkan endemic species growing at higher elevations in alpine and subalpine grasslands. In A. saxatilis the inferred phylogenies retrieved three main geographically distinct lineages. The southernmost populations exhibit an early divergence and stable in situ persistence corresponding to the species main distribution area during the LGM, whereas the central Balkan and Carpathian populations experienced more complex range dynamics triggered by Pleistocene climatic oscillations. The genetic structure of D. sylvestris revealed two groups, north-western and south-eastern, whereas species distribution modelling pointed to suitable habitats during the LGM along the whole eastern Adriatic coast with higher suitability predicted in the south compared to the north. Festuca bosniaca displayed intricate relationships with adjacent taxa of the F. varia complex, as well as a larger suitable area during LGM compared to present as suggested by distribution modelling.

### The Trapani mountains (NW Sicily), refuge for relict or highly localized endemic plants

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The Tyrrhenian coast of Sicily is flanked by a chain of mountain complexes which are the continuation of the Apennines and form the connection with the mountain ranges of North Africa, giving rise, overall, to the so-called Apennine-Maghrebi chain.

At the western end of Sicily are the mountains of Trapani which are made up of carbonate rocks originating between the Upper Jurassic-Oligocene and the Upper Triassic-Middle Jurassic. The main peaks are Mt. San Giuliano (840 m a.s.l.), Mt. Cofano (659 m a.s.l.), Mt. Speziale (913 m a.s.l.), Mt. Sparagio (1110 m a.s.l.), and Mt. Inici (1065 m a.s.l.). Although they do not reach high altitudes, they tend to stand out from the surrounding area for being isolated and for their position facing the sea. The humid currents that come from the sea allow the life of peculiar species with unique morphological characteristics; above all there is *Ptilostemon greuteri* Raimondo & Domina which has much larger leaves than the typical species of the Mediterranean flora.

The flora of this coastal mountain system is known after targeted collections by numerous botanists starting from the 19th century. Only the flora of Mt. Cofano and that of the Zingaro Nature Reserve (which includes Mt. Speziale) have been organically published. The strictly endemic taxa described in recent years and the studies still in progress demonstrate that these refuge areas deserve even greater attention from researchers. The check of the phytogeographic affinity between Mount Cofano and other calcareous reliefs of the central-northern coast of Sicily has highlighted its clear separation.

The rate of endemism of the flora of the reliefs of the mountains of Trapani is around 7%, about half of that of the entire island. This is due to the uniformity of the substrates and the reduced altitude range, while throughout the island the greater floristic diversity is expressed mainly at higher altitudes. Nonetheless, this system of mountains hosts about forty Sicilian endemic taxa, many of which are linked to the carbonate cliffs and exclusive to single mountains which for this reason qualify as micro-refuges, in some cases also of paleo-endemic species. Representative examples are *Centaurea erycina* Raimondo & Bancheva and *Silene nefelites* C. Brullo, S. Brullo, Giusso & Ilardi (Mt. San Giuliano), *Erica sicula* Guss. subsp. *sicula* (Mt. Cofano), *Hieracium cophanense* Lojac. (Mt. Cofano and Mt. Speziale), *Limonium todaroanum* Raimondo & Pignatti (Mt. Speziale), *Anthemis parlatoreana* Raimondo, Bajona, Spadaro & Di Grist., and the mentioned *Ptilostemon greuteri* (Mt. Inici).

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# Latium Flora: Distribution model of species with conservationist interest in refugia

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Refugia represent climatically and geologically stable areas that have had the function of conserving high biodiversity and high genetic diversity for a long time. It is possible to argue that the function of the refuges was not only passive, but that they contributed to increasing biodiversity through particular speciation processes and to increasing the number of endemics leading to the formation of hotspots. The concept of "refugia" was born with the need to explain the movement from north to south during the glacial periods of the populations that later returned northward during the interglacial periods. In our work, therefore, defining the pool of conservation interest species (CIS), we proceeded to an extensive bibliographic research of the historical works with consequent critical analysis in order to be able to produce an update. Then proceeded defined the different types of refugia present in the area we proceed with the distribution maps of the species selected in them. The regional flora includes a total number of taxa of 3,371 entities (23% of the overall vascular flora). The number of taxa of major conservation interest confirmed for Latium 786 entities, 766 species, 387 genera and 106 families. The most numerous genera are Carex (24), Ranunculus (15), Juncus (14), Alchemilla (11), Allium (11), Trifolium (11) and Silene (10). The most represented families are the Asteraceae (87), the Fabaceae (60), the Poaceae (53), the Brassicaceae (37), the Cyperaceae (35), the Apiaceae (34), the Caryophyllaceae (34) and the Ranunculaceae (29). Our analyzes show that the most important group to be represented with high values is that of Endemic and Sub-endemic species whose percentage increases from 6.6% to 18.7%, of Boreal species which increase from 7.1% to 11.6% and of South-European Orophytic species which instead increase from 5.6% to 9.4%. The percentage increase of these chorological types confirms that in the flora of greatest conservation interest there is an important contingent of taxa with a high biogeographical value. This largely reflects the fundamental role played by the Italian peninsula during the glacial periods of the Pleistocene and in the subsequent Holocene (postglacial). In these periods the function of shelters in the Peninsula and, in particular, in Latium was carried out in favor of populations coming from the north (boreal elements) with successive phases of ascent from the south and the start of processes of speciation by isolation. Therefore, our work of mapping of the conservation interest species in relation to the placement of the different types of refugia, has allowed us to identify in some cases to report new presences. Moreover, we identified some geographical areas with a high floristic value where there is a lack of protected areas that guarantee their adequate conservation.

# Intérêt phytogénétique et endémismes du massif du Gennargentu (Sardaigne)

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La Sardaigne, par extension la deuxième plus grande île de la Méditerranée, possède 340 entités endémiques dont 184 sont exclusives et 157 qu'elle partage principalement avec la Corse (118), ou avec la Sicile (31), l'Archipel des Baléares (10) et quelques autres avec les îles Hières et l'Archipel toscan, et les comparaisons avec l'archipel ibérique et l'Afrique du Nord ne manquent pas. L'île, contrairement à la Corse et à la Sicile, ne possède pas de plan alpin. Ce n'est que dans le système montagneux du Gennargentu, que l'on trouve des altitudes légèrement supérieures à 1 800 mètres. Le massif est caractérisée par 675 espèces indigènes, avec une nette dominance des espèces méditerranéennes, mais avec un contingent endémique de plus que100 entités dont 40 sont exclusives (ex. Aquilegia nugorensis, Astragalus genargenteus, Lamyropis microcephala, Euphrasia genargenteu, Veronica brevistyla et Rhamnus persicifolia, le seul arbre strictement endémique de l'île). Dans le Gennargentu, d'autres espèces endémiques plus répandues en Sardaigne sont Acinos sardous, Thymus catharinae, Orobanche denudata, Aquilegia barbaricina, Galium glaucophyllum, Glechoma sardoa, Iberis integerrima, Limonium morisianum et Ribes sandalioticum.

Le Gennargentu, en plus des espèces endémiques exclusives à la Sardaigne, compte 40 endémismes communs avec la Corse (ex. *Plantago sarda, Genista corsica, Euphorbia semiperfoliata, Herniaria litardierei, Lamium corsium, Odontites corsicus, Ornithogalum corsicum, Crosus minimus, Pilosella soleiroliana, Potentilla corsica, Potentilla crassinervia*) qui témoignent de l'appartenance commune au domaine floristique sarde-corse. Avec la Sicile, Le Gennargentu partage 20 espèces endémiques, parmi les quelles *Verbascun siculum, Orchis branciforti, Astragalus thermensis.* D'importantes relations floristiques sont partagées avec les autres îles mineures de la Méditerranée occidentale, l'île d'Elbe (ex. *Carlina macrocephala, Stachys glutinosa*) les îles Baléares (*Arenaria balearica, Arum pictum, Bellium bellidioides, Cymbalaria aequitriloba*) et les îles Hières (ex. *Ptilostemon casabonae*). Un autre contingent d'espèces continentales ou très rares est représenté par *Asplenium septentrionale, Blechnum spicant, Bunium corydalinum, Cerastium boissierianum, Corydalis pumila, Gentiana lutea, Ranunculus platanifolius, Valeriana montana.* Les espèces ligneuses telles que *Juniperus nana, Juniperus communis, Sorbus aucuparia, Sorbus aria, Sorbus torminalis, Amelanchier ovalis, Taxus baccata* et *Ilex aquifolium* ne sont pas moins importantes.

La composante endémique et les espèces de l'étage montagnard européen donnent à la Sardaigne, et en particulier le massif du Gennargentu, parmi les zones de refuge de la flore tertiaire, une importance considérable pour la définition des relations phytogéographiques dans la Méditerranée.

# Bryophytes in small circum-Sardinian islands: biogeographic patterns, ecology and comparison with other small circum-Italian islands

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Bryological explorations of Italy, Sardinia, Sicily, and their small islands began in the first decades of XIX century and continued still nowadays. The bryological knowledge of small Mediterranean islands is however far from complete. Within this contribution we updated bryological data of small circum-Sardinian islands. The results emerging in the present study updated the current bryo-chorological knowledge of small circum-Sardinian islands since new species were added to the Sardinian bryological flora. Moreover, the collected species were described in their functional traits by using the Ellenberg values adapted for bryophytes. More precisely, the following parameters were considered: major biome, eastern limit of distribution, light and moisture preferences, reaction to environmental acidity, occurrence in fertile to unfertile soils, and tolerance towards environmental salt and heavy metals. By this, the bryological flora of each studied island was characterized by an ecological point of view. To address ecological variability, data about circum-Sardinian islands were compared to those measured in other small Mediterranean islands.

Small islands are vulnerable niches where climate change could exert its influence. The approach implemented in the present study could be extended to other small islands to monitor future changes in their bryological flora and ecology in a scenario of a rapid climate change.

# Progress in the taxonomical and evolutionary knowledge of the aquatic liverworts *Riella*

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Riella Mont. (Riellaceae, Sphaerocarpales, Marchantiophyta) is exceptional among liverworts as it consists of ephemeral, annual species that grow submerged in seasonal ponds and lagoons. Several species can colonize brackish waters, which is unique among bryophytes. *Riella* is distributed almost world-wide in areas of rainfall seasonality, such as the Mediterranean-type climate, however, the area of most species is fragmented, and populations are commonly scattered and disjunct. Riella includes 22-29 taxa, depending on authors, with most of the species' diversity being concentrated in the western Mediterranean basin. However, this may reflect other suitable areas around the world have been underexplored. As characteristic of Sphaerocarpales, the sporophytes are enclosed within involucres. The morphology of these involucres is the basis for the recognition of two subgenera owing to their smooth/papillose (R. subg. Riella) or winged (R. subg. Trabutiella Porsild) involucres. However, ongoing phylogenetic reconstructions have challenged the monophyly of Trabutiella, supporting the repeated evolution of winged involucres in different lineages. Most taxonomically informative traits in *Riella* are derived from spores, which are variously shaped and ornamented and are essential for species identification. Recent detailed morphological and molecular studies have reassessed the taxonomy of R. helicophylla (Bory & Mont.) Mont., the type species of the genus, revealing it consisted of two species, the extremely rare R. helicophylla s. str. and R. macrocarpa (P.Allorge) Puche, Segarra-Moragues, Sabovlj., M.Infante & Heras, as a new widespread overlooked species. These results exemplify our limited knowledge on the diversity of Riella in a flagship species and point to similar results in other species groups or unexplored areas of the world, where our knowledge is incomplete or lacking. In this communication I will support this statement with some representative examples. Subgenus Riella includes most of the species' diversity of the genus, with 16-23 taxa depending on authors, and its diversity is still poorly understood. Much of this discrepancy is related to a group of monoicous taxa of the R. notarisii (Mont.) Mont. aggregate, which form a monophyletic clade, and that share overall similar morphology in vegetative and spore traits, and a distinct coriander smell. Another poorly understood taxon that probably constitutes an aggregate of pseudocryptic species is the amplitropical Riella americana Howe & Underw. This species occurs in USA and Mexico and a in single disjoint Argentinian population. Ongoing morphological and molecular studies suggest this aggregate could include four additional undescribed species thereby significantly reducing the actual range of *R. americana* and dismantling its proposed disjunct distribution.

### Mediterranean bryophytes: are there any effective conservation strategies?

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Many of the bryophytes with a Mediterranean distribution are listed as threatened in the European Red List (Hodgetts & al. 2019: https://doi.org/10.2305/ IUCN.CH.2019.ERL.2.en). Threats include habitat destruction and degradation, spread of invasive species and climate change. However, many species suffer from insufficient information about their distribution and ecology, many have an ephemeral life strategy, and many are sporadic and unpredictable in occurrence. This results in some difficult challenges for those involved in their conservation.

The Mediterranean region is on the front line of climate change, with increasingly unpredictable weather conditions leading to prolonged droughts, wildfires and floods, while simultaneously there is increasing pressure on land and resources from issues related to tourism, building construction and farming.

For effective conservation, the requirements of Mediterranean bryophytes must be considered along with those of other taxonomic groups and their conservation integrated into wider initiatives. A number of species are chosen as case studies to highlight their current status, particular problems associated with their conservation, and how their conservation might best be addressed.

# Decoding the determinants of the abundance and distribution of Mediterranean bryophytes

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Understanding how micro and macro climate influence bryophyte species abundance and distribution has been at the center of diversity and biogeography studies for decades. Numerous researchers have focused on small-scale influences such as microclimate to explain species presence and abundance in specific locations while others have pursued the same question by examining larger scale factors. These efforts have brought valuable insights, yet the relative importance of small vs. large scale factors remains elusive due to a lack of integrative studies that consider both scales.

Recently, a more integrative approach has emerged, combining both microclimatic and macroclimatic characteristics to analyze species abundance and distribution. These studies have underscored the critical importance of considering the scale in our understanding of species distribution.

Furthermore, these studies have highlighted differences in the factors affecting bryophyte distribution across climate zones. Specifically, the roles of micro and macroclimatic factors across scales differ fundamentally between Mediterranean and Eurosiberian climates.

This talk aims to delve further into these recent findings, fostering a more nuanced understanding of bryophyte distribution across scales. The focus will be specifically on Mediterranean climates, an area of particular interest due to the unique interplay of microand macroclimatic factors influencing bryophyte distribution.

# In search of strictly Mediterranean lichens: contribution of collection based approach to characterization of European Mediterranean

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Current species distributin patterns are closely determined by ecological and evolutionary correlates. Mediterranean regions have featured long-term climatic stability since the end of the Miocene. This stability is also observed in saxicolous habitats, which support a diverse range of lichens that are assumed to be less affected by the future climate. Building on our previous research in phylogeny, phylogeography, biogeography as well as ecology of European Solenopsora congeners (Leprocaulaceae), we conducted collection-based research to map the distribution of these saxicolous lichens. We utilized a comprehensive dataset from 40 lichenological collections covering the Mediterranean basin, adjacent Atlantic regions, and continental areas (BM, BP, BRA, BR-LICH, BRNU, BUCM, CBFS, CLU, FH, FI, G, GZU, H, Herb. I. Pišút, Herb. J. Malíček, herb. J. Vondrák, Herb. Linda in Arcadia, herb. P. van den Boom, herb. Z. Palice, KHER, KRAM-L, LD, LISU, M, MAF-Lich, MSC, O-L, PRA, PRC, PRM, SAV, S-L, TO, TSB, UBC, UPSL, VAB-LICH, VER, W, and ZA). Using WorldClim data, we analysed the climatic envelopes of the studied taxa and their correlation to bioclimatic zones. We constructed predictive habitat suitability models, which indicate the potential distribution of suitable sites. By constructing predictive habitat suitability models, we identified potential distribution sites for these lichens. We also reconstructed habitat suitability under past climatic scenarios and projected it for 2050 and 2070. We determined ecological niches of the studied taxa in Europe and the key ecological factors shaping their environmental niches, considering both climatic and non-climatic drivers. Four of the studied taxa can be considered strictly Mediterranean.

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# Threatened Liverworts of Italian bryoflora: analisys of biological and ecological characters

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Considering of the need to have an updated view of the conservation status of the Italian bryophytes flora, the "Gruppo per la Briologia" of the Italian Botanical Society has recently started compiling the new red list, of which the first part has just been published, relating to the Liverworts and Hornworts. Twenty years after the first "Red Book of Italian bryophytes", the new red list follows the criteria and guidelines provided by IUCN and reports the results of the evaluation of a total of 306 taxa so far known for Italy, on the basis of what is reported in the last check list of this territory. Taxa that are referred to one of the risk categories are now being analyzed from an ecological and biological point of view in order to provide useful data for better understanding and protect this most fragile component of the Italian bryoflora, also considering the opportunity to have data on populations of these taxa with reference, for example, to the reproductive capacity or to the ecological features. Furthermore, the analysis will also concern taxa assessed as Data Deficient, for many of which there is often insufficient knowledge of their distribution and targeted field research would be necessary, for which it may be useful to have data on the specific ecological traits of the taxa to be researched.

### Biodiversity of fungi in freshwater ecosystems of Italy

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The ubiquitous group of freshwater fungi includes any species that spend their entire lifecycle, or part of it, in freshwater habitats (ponds, pools, lakes, peat swaps, wetlands, rivers, streams, artificial reservoirs, etc...) colonizing submerged organic matter such as leaves, wood, and stems of plant. Currently, about 4,000 species of freshwater fungal species are known, but considering that many freshwater habitats are still unexplored, this number is probably underestimated. Different taxonomic groups can be recognized and separated according to their morphology and ecology: freshwater ascomycetes, freshwater hyphomycetes ("Ingoldian fungi", aero-aquatic hyphomycetes, terrestrial-aquatic hyphomycetes, submerged-aquatic hyphomycetes), freshwater basidiomycetes, coelomycetes, zygomycetes, microsporidia, and zoosporic fungi. They belong to 13 phyla and among them Ascomycota are the most numerous (2,968 species). Data collection about freshwater fungi derives mostly from temperate areas of Asia, Australia, North America, and Europe but a very few data are reported about their presence in freshwater ecosystems of Italy. In this survey, the current knowledge of fungal diversity in freshwater ecosystems is analyzed. It is appropriate to specify that yeasts, Chromista, and lichens will not be taken into consideration in this study. From 1888 to 2023 only 16 studies are reported about the presence of freshwater fungi in Italy. Fourteen of them examine Ascomycota and only two are referred to Basidiomycota. Most of these studies were carried out in Northern and Central Italy (Lombardia, Piemonte, Veneto, Umbria, and Lazio) where the biggest Italian lake and rivers are located, while data about southern Italian region and islands are basically absent. The list presented in this survey reports 130 species of Ascomycota belonging to Chytridiomycetes, Dothideomycetes, Eurotiomycetes, Leotiomycetes (the most numerous), Laboulbeniomycetes, Pezizomycetes, and Sordariomycetes classes and about 150 species of Basidiomycota belonging to Agaricomycetes. While Ascomycota are often isolated directly from water or submerged organic matter, Basidiomycota are always report associated with litter and part of trees or vegetation along freshwater habitats. Considering how many Italian freshwater habitats are still unexplored and considering the ecological importance of freshwater fungi further studies are needed in order to expand the knowledge of this group of fungi and protect their biodiversity.

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#### Balkan diploids and polyploids in Cardamine (Brassicaceae)

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The genus *Cardamine* is a cosmopolitan genus, comprising almost 280 species (Marhold et al. 2021+, www.cardamine.sav.sk). In the Balkan Peninsula it is represented mostly by diploids whereas polyploids occur there only exceptionally (species previously classified within the genus *Dentaria*, *C. flexuosa* and few other taxa).

The complex of taxa related to *C. acris*, *C. amara* and *C. pratensis* that is dealt with in this contribution include mostly endemics (*C. amara* subsp. *balcanica*, *C. acris* subsp. *acris*, *C. acris* subsp. *vardousiae*, *C. acris* subsp. *pindicola*, *C. barbaraeoides*, *C. penzesii*, *C. rivularis*) but also more widespread European taxa reaching their southeastern distribution in the Balkans (*C. matthioli*, *C. amara* subsp. *amara*, *C. amara* subsp. *opicii*). Apart from tetraploid records for C. barbaraeoides and triploid plants reported for *C. rivularis* and *C. ×rhodopaea* (*C. rivularis* × C. matthioli), these taxa were known as diploids.

In order to resolve the phylogenetic relationships among the above-mentioned taxa and particularly the origin of tetraploid *C. barbaraeoides* endemic to the Southern Pindos Mts. (Greece) we used particularly target enrichment with genome skimming (Hyb-Seq) and genomic in situ hybridization (GISH). We obtained a robust phylogenetic reconstruction for diploids based on 1,168 low-copy nuclear genes, which suggested both allopatric and ecological speciation events. In addition, cases of plastid–nuclear discordance, in agreement with divergent nuclear ribosomal DNA (nrDNA) copy variants in some species, indicated traces of interspecific gene flow. An allopolyploid origin was inferred for *C. barbaraeoides*, which highlights the role of mountains in the Balkan Peninsula both as refugia and melting pots favouring species contacts and polyploid evolution in response to Pleistocene climate-induced range dynamics.

Very complex genome size patterns were found in the Bulgarian range of *C. rivularis*, where apparently both unreduced gamete formation and hybridization with *C. matthioli* and most likely also with *C. amara* subsp. *balcanica* have resulted in a mixture of different autoand allotriploids persisting along with diploids. The substantial genome size variation revealed within as well as among populations may be due to multiple hybrid formation and even crossing between triploids of different origins. The genus *Anacyclus (Anthemideae, Asteraceae)* as a model for research on diploid hybrid lineages evolution

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Species or lineages of hybrid origin are relatively frequent in plants. In many cases, a change in the number of chromosomes, the current presence of the parental lineages and the appearance of intermediate states of characters in these species clearly evidence their hybrid origin. However, in other cases in which there are no changes in the number of chromosomes and the morphological characters are similar to that of one of the parents, the hybridization process may go unnoticed or be difficult to elucidate. In these cases, each of the pieces of evidence alone may not be sufficient and it is crucial to collect evidence of various kinds in order to prove such hypotheses.

Anacyclus is a small genus (<10 species) of diploid (n = 9) mainly annual herbs that occupy anthropic areas, whose centre of diversity is located in NW Africa, widespread in the Iberian Peninsula and scattered throughout the Mediterranean. The diversity in their floral characters and the intermediate states observed in some of them in phenotypically mixed natural populations has been interpreted as a consequence of interspecific hybridization. In addition, the hybrid origin of some of its species has been hypothesised for years. For more than a decade several research groups have been interested in this genus, carrying out phylogenetic, genetic, cytogenetic and genomic analyses of natural populations, field experiments to estimate fitness, and experiments with artificial crosses of several hybrid generations to understand the evolution of this group of species. Among the most relevant results we can highlight a new circumscription of the genus, the role of chromosome rearrangements in its evolution, the support of the hybrid origin of at least one of its species, the discovering of a new cryptic lineage, the need of a revision of the diagnosis characters for species delimitation, and finally the species concept in this group and in similar plant systems. We present here a synthesis and discussion of these results that demonstrate the great complexity in the study of diploid hybrid lineages and the relevant role of hybridization for the evolution of this group of plants.

# Morphometric tools to test species delimitation: the case of Armeria (*Plumbaginaceae*) in central Mediterranean area

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Morphological characters have long been used in taxonomy to differentiate species. Despite advances in phylogenomics, morphology remains crucial in botany. Taxonomists subjectively select characters, trying to disregard interindividual variability influenced by genetics, ecology, and development to document differences in keys or descriptions. However, the complexity of nature and unique biological adaptations in plants can challenge taxonomists, leading to biased decisions and incorrect assignment of interindividual variability to a certain taxonomic rank. Morphometry, established as a science by R.E. Blackith in 1957, allows to perform more rigorous morphological comparisons. This approach has proven effective in difficult genera such *Armeria (Plumbaginaceae*), where classical taxonomy may struggle due to variation, introgression, and hybridization.

In the last two decades, Principal Component Analysis and Linear Discriminant Analysis paired with boxplots dominated morphometric studies in plants. However, these methods are limited due to assumptions that are rarely met in practice. Indeed, mixed data, non-multivariate normal, and high-dimensional datasets are common in morphometry. Nowadays, machine learning (ML) has great potential in advancing morphometry, enabling more accurate species circumscription and identification at the end of the taxonomic process. In case of mixed data, proper distances or encoder for categorical data must be chosen and a proper data preprocessing is required. Regularized supervised classification algorithms like Regularized Discriminant Analysis (RDA) and Elastic-Net Logistic Regression (EN LR) better handle high-dimensional numerical datasets. In particular, EN LR proved to be effective in the delimitation between A. denticulata and A. saviana, two central Italian endemics. When there are many categorical data, Random Forest can be employed for data visualization, classification and during the construction of an identification key. For small sample sizes, nonparametric models like kNN work well as they require minimal assumptions, have no parameters to penalize, and accurately estimate decision boundaries between taxa. Due to the limited number of individuals available for the taxa of Armeria from Sardinia and Corsica, kNN proved to be well-suited for their morphometric study. Kernel density estimators could be paired with boxplots, since the latter may mask underling bimodal distributions.

An intriguing future development in morphometry concerns semi-supervised ML, i. e. the automated identification by classifying unknown objects based on proximity to labelled objects in the morphospace. This approach accounts for noise, errors, and variability. Semi-supervised ML trained on morphometric data can accurately identify unknown individuals using statistically selected measurements, contrasting with subjective character selection in classical taxonomy and equal weighting in classical numerical taxonomy.

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### Disentangling relationships in different Mediterranean *Euphorbia* lineages (*Euphorbiaceae*) using integrative approach

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*Euphorbia* is one of the largest genera of flowering plants including more than 2150 species. Most species in Europe belong to *Euphorbia* subgen. *Esula*, which is sister to three other subgenera. It comprises roughly 480 species and represents the most significant radiation of the genus in temperate areas of the Old World, with high diversity in the Mediterranean Basin, which is a hotspot of European biodiversity. I will present the outcomes of past and ongoing studies on diversification of different Mediterranean *Euphorbia* species, based on DNA sequencing, including next generation RAD sequencing, AFLP fingerprinting, relative genome size measurements, chromosome number estimations and morphological analyses. More specifically, I will present (1) the evolutionary pathways leading to the origin of a recently described allotetraploid *Euphorbia ligustica* endemic to Liguria, (2) phylogeographic relationships among the Apennine-Balkan *E. adriatica* and *E. japygica*, as well as (3) diversification patterns within the Mediterranean species of *Euphorbia* sect. *Patellares* that were in the past considered conspecific or closely related to *E. amygdaloides*, namely *E. durandoi, E. heldreichii, E. meuseli, E. orjeni* and *E. semiperfoliata*.

### Towards a resolved phylogeny for a new classification of Crocus (Iridaceae)

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The genus *Crocus*, comprising about 240 species are distributed from western Europe and northwestern Africa to western China. The genus requires a taxonomic revision as several infrageneric and even intraspecific units were shown to be para- or polyphyletic. Additionally, over 60 new species were described recently but, in most cases, not assigned to existing taxonomical groups. Therefore, this study was aimed to resolve the relationships within the genus for a better understanding of the genus history and as a base for the definition of new taxonomical units, which reflect phylogenetic relationships.

Through a comprehensive analysis using whole genome shotgun sequencing (200 samples) as well as three chloroplast and 16 nuclear markers (500 samples) the backbone of the genus could finally be resolved showing division into four main lineages. These main lineages diverged within the first 2 million years after *Crocus* originated (17.7 - 21.9 million years ago). Further diversification took place shortly afterwards in the Oligocene (about 12-15 million years ago), especially in the eastern Mediterranean area. From there, the western Mediterranean area was colonized. Most of today's existing species originated within the last five million years.

Relationships within three of the four main lineages could be well resolved. However, there were strong incongruences between markers within the fourth main group (section *Nudiscapus* s. str.) most probably caused by hybridization.

The detection of hybridization events is sometimes complicated by fast diploidization resulting in the loss of parental alleles even in younger hybrids. In such cases, a clear identification of hybridization events and morphological characterization of diploids and alloploids was possible by linking molecular-phylogenetic results, genome-wide genotyping-by-sequencing (GBS) data, chromosome counts and genome-size measurements with morphology. Identification of older hybridization events in *Crocus* is often more difficult. Extensive chromosomal rearrangements are very common in the genus resulting in highly variable chromosome numbers and genome sizes, which do not necessarily correlate with each other. Therefore, hybridization and polyploidization events cannot be derived from chromosome counts and/or genome sizes. In such cases, hybridizations are often only detectable by incongruence between different markers. Generally, hybridizations events were more prominent in complexes with confusing and conflicting taxonomy. In addition to generally resolving the phylogeny of the genus, the identification of hybridization events helped to evaluate morphological characters and character combination for a better definition and recognition of infrageneric taxonomical units.

# From error-prone Nanopore reads to valuable insights: AFLP-based genome reduction and the SLANG pipeline for multi-locus analysis in biosystematic research

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Next-Generation Sequencing (NGS) techniques, such as Restriction-site associated DNAsequencing (RADseq) and Genotyping-by-Sequencing (GBS), are powerful and cost-effective genome-reduction methods used in biosystematic studies for generating anonymous multi-locus data. They have proven valuable for genotyping, phylogenetics, and species delimitation, especially in taxonomically challenging groups. Some well-developed bioinformatic pipelines are available for analysis of high-quality, short-read Illumina data, with IPYRAD and STACKS being the most prominent ones for the *de novo* reconstruction of loci from single reads and the subsequent orthology estimation and single-nucleotide polymorphism (SNP) calling through grouping reads and loci by similarity.

Over the last few years, Nanopore sequencing, particularly using the small-sized MinION system (Oxford Nanopore Technologies), has gained popularity due to its low initial cost compared to Illumina sequencers. The MinION's affordability, reusable flow cells, and smaller capacity flow cells (Flongle) make NGS feasible for smaller molecular-systematic laboratories.

However, unlike with high-quality Illumina sequence data, there is a lack of bioinformatic tools specifically tailored to the analysis of error-prone Nanopore multi-locus read data. To address this gap, we developed SLANG (Simple Long-read loci Assembly of Nanopore data for Genotyping), a Python-based pipeline for Nanopore data analysis. SLANG clusters all reads of operational taxonomic units (OTUs), enabling simultaneous *de novo* assembly and orthologization of loci. SNPs are automatically extracted by mapping reads to cluster consensus sequences and can be further filtered according to individual needs.

We applied SLANG to several taxonomic studies as proof-of-concept. In the central European *Senecio nemorensis*-group (Senecioneae, Compositae), including *S. hercynicus* Herborg, *S. ovatus* (G. Gaertn., B. Mey. & Scherb.) Willd., and *S. germanicus* Wallr., SLANG successfully differentiated the species. SLANG also aided in genetically characterizing the new species *Adonis fucensis* F.Conti & Bartolucci (A. sect. Adonanthe, Ranunculaceae) and delimiting *Pedicularis rostratospicata* subsp. *marsica* F.Conti & Bartolucci (Orobanchaceae) as a new subspecies.

To generate multi-locus datasets, we used the well-known AFLP-based genome reduction with selective bases influencing locus numbers and sequencing depth, which complements the scalability of Nanopore sequencing. Owing to the growing interest for utilizing the potential of herbarium specimens, we additionally demonstrate the usability of AFLPs for herbarium specimens in the mentioned studies. Overall, AFLPs in combination with the newly developed SLANG pipeline proved to be a valuable alternative to conventional NGS and AFLPs.

### Exploring the phylogenetic relationships in *Santolina* (*Asteraceae*): a taxonomically complex genus endemic to the Mediterranean Basin

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Rapid divergence, introgressive hybridisation and polyploidization often result in taxonomically and evolutionary complex groups with weakly geographically and/or morphologically defined species. In these groups, the difficulty to classify species in stable and coherent taxa strongly affects the implementation of conservation measures for threatened, rare or endemic evolutionary significant units, thus, to preserve the processes that lead to the generation of biodiversity. The genus Santolina L. (Asteraceae, Anthemidae), endemic to the western part of the Mediterranean basin, has a long taxonomic history since Linnaeus (1753) described the genus. According to a recent taxonomic revision, the whole genus comprises roughly 30 taxa, most of which are divided in two morphological complexes: the rosmarinifolia one, which includes eleven taxa endemic to Iberian Peninsula and North Africa and was subject of extensive systematic and phylogenetic analysis, and the *chamaecyparissus* one, which includes fourteen taxa mainly narrow endemics occurring in Spain, France, and Italy. In addition, four taxa are not included in either complex. In this study, for the first time we presented a genome-wide phylogenetic analysis of the whole genus based on RADseq. We also investigated diversity structure by computing a co-ancestry matrix using RADpainter software. To assess evidence of introgression between species, we used Dsuite package performing the ABBA-BABA test. Our phylogeny corroborates the results of previous morphological analyses, confirming the existence of two main clades: the rosmarinifolia clade, which is monophyletic; and the *chamaecyparissus* clade which is monophyletic if the allopolyploid S. villosa is excluded. Santolina villosa (chamaecyparissus complex) shares common ancestor with species of S. rosmarinifolia complex. Taxa belonging to rosmarinifolia clade shown high level of co-ancestry supporting the phylogenetic closeness of these species. Taxa of chamaecyparissus clade are genetically more heterogeneous and form multiple subclusters. The ABBA-BABA tests found that the degree of introgression varies among taxa, being generally high between taxa of the chamaecyparissus clade and the rosmarinifolia clade. The introgression between the two early diverging lineages (i.e., S. rosmarinifolia and S. chamaecyparissus clades) was probably a crucial factor supplying the genetic diversity required for the radiation of *Santolina* lineages, particularly in the sympatric species of the rosmarinifolia clade. In conclusion, we build the first evolutionary hypothesis for Santolina chamaecyparissus complex, which yields a much-increased understanding of phylogenetic relationships in this group. Taken together our results set the stage for further investigations of the biogeographic history of this circum-Mediterranean group.

# The evolutionary history of junipers (*Juniperus* sect. *Juniperus*) based on genomic (GBS) analysis

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Junipers are coniferous trees or shrubs belonging to the cypress family and distributed across the Northern Hemisphere. Depending on the taxonomic treatment, Juniperus L. is recognized to encompass 53 to 76 species, of which 7 to 14 are included into Juniperus sect. Juniperus L. Previous phylogenetic studies support the idea that J. sect. Juniperus constitutes a natural evolutionary group, which includes plants restricted to the Old World (with the exception of the circumboreal J. communis L.). However, little is known about the phylogenetic relationships within the section due to the low resolution of the genetic markers used up to date. To resolve this puzzle, we presented a reduce-representation genome (GBS) strategy including samples from all the species considered in J. sect. Juniperus. To compare the performance of different assembly methodologies, we run both denovo and reference (using Cupressus sempervirens L. genome as reference) assemblies testing several parameters in order to get the most informative alignments. The referenced dataset includes a higher number of Pis (parsimony informative sites) and recovered loci, but it also presents the highest percentage of missing data. The phylogenetic inference method used for tree reconstruction, based on either a concatenated supermatrix or a summary gene tree methods, were performed for both assembly strategies (reference or denovo). The results conclude that J. sect. Juniperus comprises two natural lineages: one including species related to the J. communis complex, which includes the Far East distributed representatives, and another group including J. oxycedrus L. (prickly juniper) and allies, with a narrower distribution encompassing the Mediterranean area and part of the Macaronesian archipelagos. We discuss the taxonomic status of various members of the latter lineage, confirming the taxonomic species-level range of several taxa traditionally recognized as subordinate to J. oxycedrus. These taxa include J. badia (H.Gay) Rivas Mart., Molero Mesa, Marfil & G.Benítez, J. deltoides R.P.Adams, J. maderensis (Menezes) R.P.Adams, J. navicularis Gand, and J. willkommii Antoine. Additionally, we present a dated phylogeny to track diversification rates within J. sect. Juniperus.

### Tackling taxonomic inflation: biosystematics studies of the *Dianthus* virgineus complex (*Caryophyllaceae*) in the Central Mediterranean

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Circumscribing species according to qualitative observations can produce taxonomic inflation. Over-estimating the number of taxa in a group may have two main negative consequences: undermining the credibility of systematics within the scientific community and biasing prioritization of resources in conservation projects. Plant taxonomists working in the Mediterranean area often deal with taxonomically doubtful and complex groups of narrowly related species.

For instance, the *Dianthus virgineus* L. complex includes many described wild carnation species, mainly distributed in the central Mediterranean (Sardinia, Sicily, and central-southern Italy), where 21 taxa have been recognized mostly based on morphology. Testing species support with multiple lines of evidence could provide a solution to possible taxonomic inflation. Accordingly, we are following integrative taxonomic approaches to test all central Mediterranean taxa within the *D. virgineus* complex. We collected morphometric, cytogenetic, and genomic (ddRAD-seq) data from over 100 populations belonging to all the 21 central Mediterranean taxa.

Multivariate morphometric analyses highlight a difficult recognition of currently accepted taxa in the morphospace constructed using qualitative and quantitative variables. Morphological data support the distinction of a few population groups. In general, populations distributed from southern France to peninsular Italy show shorter calyx teeth, epicalyx scales and mucros with respect to populations from Sicily and Corso-Sardinian system (including Tuscan Archipelago). Although the chromosome number is constant (2n = 2x = 30) among and within the studied taxa, relative genome size (RGS) is higher in populations. The characterization of thousands of genome-wide SNPs revealed a genetic structure that parallels geography rather than current taxonomy. All the studied populations from the central Mediterranean belong to a unique genetic lineage. Moreover, two main genetic clines were retrieved: one from southern France down to southern peninsular Italy, the other one from Italian Peninsula to Tuscan Archipelago, Corsica, and Sardinia. Sicilian and Sardinian populations are related through those from Tunisia.

Multiple lines of evidence support the presence of a lower number of taxa than currently accepted, and taxonomic changes will be required to obtain a coherent, more useful, classification system of this group.

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# Diversity of the Italian populations of *Onopordum tauricum* (*Asteraceae*) as revealed by ddRAD sequencing and morphometric data analysis

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*Onopordum tauricum* Willd. is a species occurring in both Mediterranean and temperate climates and its native range is represented by south-east and the Near East, having its centre of dispersion in the Black Sea area. It is regarded as a noxious species and it was accidentally introduced to Australia and the United States. The Italian range of this species is extremely fragmented and it is characterised by small populations with a localized distribution. It occurs in synanthropic habitats with the presence of domestic animals, especially of sheep, on soils rich in nitrate and organic matter.

The Tauricum thistle was one of the plant involved in the European Project PRIMA "Valorisation of thistle-curdled CHEESES in MEDiterranean marginal areas" (acronym "VEG-GIE-MED-CHEESES") aimed at finding suitable plants for the production of vegetable rennet to be used in the production of local cheeses.

In order to be successful with the cultivation of this wild species, a deep knowledge of the species of interest is necessary. Indeed, the knowledge of wild plants in terms of their biology is rather poor and deepening the knowledge of their genetics and genomic traits can be regarded as the first step to evaluate possible strategies for their domestication, also with reference to the geographic origin of the individuals to be cultivated and the area where their cultivation will be carried out. Genetic improvement of wild plants for cultivation purposes could be achieved through a clear understanding of plant's ecology and the extent of variability of wild populations, including genotypes that could hold a great potentiality for adaptation to different ecological conditions. Therefore, the identification of "ecotypes" could be important to choose which germoplasm should be used in a certain geographic area. Moreover, the Italian populations occurs in the western limit of the native distribution range of the species so that the different climatic and environmental conditions could represent a strong pressure toward the adaptation and the appearance of specific characters.

With this study, the genetic and morphological diversity of the six Italian populations was evaluated through ddRAD-seq methodologies and the measurement of 37 different morphological traits; furthermore, the phylogenetic position of *Onopordum tauricum* among the subtribe of Onopordineae was clarified and the genetic structure of the Italian populations was described in order to assess if they can be recognized as different ecotypes according to the different ecological conditions of their habitats.

# The challenge of conservation and restoration of macroalgal forests with special attention to the Mediterranean

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The field of marine habitat restoration has developed rapidly in recent years and is likely to accelerate with the UN Decade of Restoration (2021-2030). Despite a relatively long history in some countries, the science and practice of marine forest restoration in the Mediterranean is still in its infancy and there is much to learn from failures and successes. Furthermore, the field of macroalgal restoration lags behind other marine ecosystems, with fewer projects and smaller restoration efforts.

Recently, growing awareness and concern about the increasing threats and observed declines has led to several publications with recommendations for the restoration of *Cystoseira s. l.* 

To some extent, biological, environmental and logistical challenges have been overcome and lessons learned can help in selecting the best sites, species and protocols for restoration. While climate change makes restoration urgent, it also limits its feasibility. Both natural and restored populations face the same threats (e.g., thermal anomalies, storm surges) and it may no longer be possible or advisable to restore the same species or population in an area where it previously occurred.

An important challenge is to address the often difficult goal of scaling the impact of restoration to the scale of forest loss. Effective scaling requires consideration of spatial and temporal variability in environmental factors (e.g., nutrients, temperature, local oceanography), stressors and connectivity, as well as consideration of cost-effectiveness, permitting constraints and logistical support needs to ensure the feasibility and success of restoration.

### What's new in *Acrosymphytales* (*Rhodophyta*)? The case of Mediterranean *Acrosymphyton purpuriferum*

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The comprehensive study of Mediterranean Acrosymphyton purpuriferum (J.Agardh) G.Sjöstedt (Acrosymphytaceae, Acrosymphytales, Rhodophyta) resulted in significant amendments to the description of various aspects of its postfertilization stages. The findings revealed that the development of the carpogonial fusion cell is mediated by conjunctor cells, which challenges the previously documented fusion process. Additionally, the study provided evidence for the existence of primary auxiliary fusion cells prior to diploidization, which had neither been reported nor illustrated before. On the other hand, the Acrosymphytales currently includes three families: Acrosymphytaceae, Schimmelmanniaceae, and Acrothesauraceae. Traditionally, the Acrosymphytaceae have been regarded as typically non-procarpic. However, this study has shown that A. purpuriferum, the type species of the family, may occasionally exhibit procarpic behavior with gonimoblasts developing from the carpogonial fusion complex. On the other hand, Schimmelmanniaceae are entirely procarpic, whereas Acrothesaurum gemellifilum Kraft & G.W.Saunders, the type and only species within Acrothesauraceae, can display both procarpic and non-procarpic behaviors. In this presentation, the coexistence of both procarpic and non-procarpic behaviors at various taxonomic levels will be discussed, with a focus on species exhibiting both behaviors. The evidence suggests that procarpic behavior is likely secondarily derived, as previously proposed by other authors.

# Understanding the invisible - population and community ecology of algae from Mediterranean streams on two continents

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Mediterranean biomes on two continents are unified by plenty of light and limited variable precipitation but also are influenced by surface and groundwater hydrology, chemistry, and anthropogenic influences. Algal populations are expected to adapt to those often-harsh conditions but the community similarities through time and across geographic regions are unknown. In long term studies with many samples, original enumerations and identifications are completed by different individuals through numerous taxonomic changes and updates. The goals of this research were to review existing documentation from Mediterranean springs and streams in Cyprus and Southern California, updating taxonomy to current concepts and inferring ecological community dynamics. Detailed documentation was reviewed, and taxonomic precision was applied to strengthen ecological inferences and address concepts in biogeography and evolutionary development. Taxa like Cymbellonitzschia diluviana Hustedt and Sellaphora stroemii (Hustedt) H. Kobayasi were common in occurrence but usually in low abundance in both locations. Common abundant representatives of Nitzschia (N. archibaldii Lange-Bertalot, N. palea (Kützing) W. Smith) and Ulnaria (U. monodii (Guermeur) Cantonati et Lange-Bertalot) differed between the two locations. The initial colonizers Achnanthidium tepidaricola Van de Vijver et M. De Haan and Crenotia rumrichorum (Lange-Bertalot) Wojtal from high mineral-content waters were documented in Cyprus only and varied through time. For Southern California, monoraphid diatoms Gogorevia exilis (Kützing) Kulik. & Kociolek, Planothidium amphibium C.E. Wetzel, L. Ector & L. Pfister, P. pericavum (J.R. Carter) Lange-Bertalot and/or P. engelbrechtii (Cholnoky) Round & L. Bukhtiyarova were common, abundant, and unique. Lotic diatom communities from the two geographically distant areas with Mediterranean climate on two continents differed through time and based on location but were not dominated by high nutrient ubiquitous diatom populations.

### New taxonomic and ecological data on new-to-science, rare and insufficiently known *Nitzschia* species (*Bacillariophyta*) from the Island of Cyprus

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While studying the diatom communities in perennial and intermittent streams in the waterstressed Island of Cyprus, we came across a number of intriguing and unrecognized diatom species belonging to the taxonomically complex genus Nitzschia. This contribution focused on three *Nitzschia* species that were either new to science, geographically limited, or poorlyknown. Nitzschia saprotolerans sp. nov., morphologically superficially resembling short and stocky morphotypes of Nitzschia fonticola, is described as species new to science on the basis of an integrated molecular, morphological, and ecological approach. More in-depth ecological information on Nitzschia pseudalpina E. Reichardt, a species newly described in 2018, was given based on extensive hydrological and chemical data collected over the sampling years (2011, 2018, 2019, 2020), as well as diatom count datasets. Lastly, some more observations on the state-of-the-art taxonomy and distribution of the Sardinian morphotype/variant of Nitzschia transtagensis E.A.Morales, Novais, C.E.Wetzel, M.M. Morais & Ector 2020 were provided. Conducting such a comprehensive taxonomic, molecular and ecological investigation on diatoms in these Mediterranean streams, which are currently experiencing fast and extensive climate change, is highly relevant for utilizing these organisms as effective indicators of environmental status and human pressures.

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## Ecology, conservation, and taxonomy of diatoms in Mediterranean streams and springs in the face of climate change

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Mediterranean streams are naturally highly stressed environments mainly due to the ample seasonal and inter-annual fluctuations in water quantity. The large human population coupled with low rainfall (which will be exacerbated by climate change), combine to create a pressure on both quantity and quality of water resources. This, in turn, will affect the ability of Mediterranean freshwaters to deliver ecosystem services and have implications for sustainable growth.

Mediterranean spring ecosystems are unique habitats that support a remarkable  $\gamma$ -diversity and provide important ecological functions and ecosystem services. A wide array of species benefits from the stability of these ecosystems, particularly during dry periods. However, changes in temperature, evapotranspiration and precipitation patterns can alter the water balance and chemistry of these fragile systems. Eutrophication due to agricultural practices and emergent pollutants are also a growing concern for the preservation of Mediterranean spring biodiversity.

Even a tentative application of a Red-List approach underlined the overwhelming importance of hydrology-related variables in determining diatom species distribution and community ecological attributes in the water-stressed Island of Cyprus. Somewhat unexpectedly, both species from threat categories of the diatom Red List for Central Europe (2018) and species one might predict would be included in such categories in a possible future Red List tailored for Cyprus occurred more frequently and were more relevant in intermittent streams.

Sensitive, characteristic, endangered diatom species are found in Mediterranean stream and spring sites with high ecological integrity. Examples of species new to science described from these habitats are used to discuss issues in diatom taxonomy in Mediterranean freshwaters.

Pollution and contamination of various types typically causes a simplification and generalization of diatom assemblages, with dominance of a reduced number of pollution-tolerant species and, in some cases, accompanied by an increase in deformed specimens. Diatom data collected from Cypriot streams in 2010-11 and 2020-22, respectively, were used to check if a banalization of communities might have occurred in ten years of marked climate change in the Eastern Mediterranean.

In-depth knowledge to foster conservation of Mediterranean stream and spring diatoms is of pivotal importance in these critical decades of environmental change.

## Biodiversity associated with Mediterranean fucalean forests and mechanisms regulating it: an assessment of the current knowledge

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Forests formed by fucalean brown algae of the *Cystoseira s. l.* complex (genera *Cystoseira, Ericaria* and *Gongolaria*) are a typical feature of shallow Mediterranean seascapes. These seaweeds play a key role in shaping biodiversity on shallow rocky bottoms; due to their highly branched morphology, they increase tridimensionality and structural complexity of benthic assemblages, providing a high diversity of habitats for many sessile and mobile organisms. Studies concerning the biodiversity of *Cystoseira s. l.* forests have largely focused on the associated macroalgal and invertebrate assemblages. Inventories presented in these studies usually consist of floristic and/or faunal lists based on traditional morphological identifications. The majority of them was carried out at local spatial scales, *i. e.* an individual site or a few sites in a geographic area; *Ericaria crinita* is the species for which the largest sampling effort and geographical coverage is currently available. To date, information concerning the microalgal assemblages associated with *Cystoseira s. l.* is also extremely limited, despite of the role plaid by this component on the well-being of the macroalgae.

Several hundreds of macroalgal and invertebrate taxa have been reported in association with *Cystoseira s. l.* species (up to 597 taxa recorded for *Ericaria corniculata* and *E. brachy-carpa*), with substantial differences among species. For individual species, significant variation in associated biodiversity has been detected at regional scale. Experimental studies demonstrated that loss of *Cystoseira s. l.* canopies involves major changes in the structure of benthic assemblages. Density of the canopy and size of *Cystoseira s. l.* patches directly affect the number of associated benthic taxa.

Future investigations should increase the number of *Cystoseira s. l.* taxa sampled and extend the geographic coverage (especially to eastern Mediterranean regions). Molecular data such as eDNA sequences should be incorporated in future studies. Finally, the microbiomes associated to *Cystoseira s. l.* forests should be investigated with more detail.

## DNA barcodes as permanent labelling in floristic and systematic studies of macroalgae

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Macroalgae are a heterogeneous assemblage of photosynthetic organisms spread in the eukaryotic tree of life. Regardless their huge variety of morphologies, anatomical constructs and genetic structures, they share the common attitude to display a high degree of phenotypic plasticity as well as morphological convergence, which make their taxonomic identification a difficult and tricky task.

As a consequence, DNA identifications became more and more relevant in phycological studies since the end of the last century. As opposed to morphological traits, DNA sequences proved to be more useful for uniting biological specimens into genetic groups as a first step to assigning them to species and genera. Even if phylogenetics and systematics should explore all available methodologies and genetic markers in the boundless framework of scientific research, a standard technique gained relevance in the last decades in order to build a DNA library of life, with data sharable among research laboratories worldwide, and among them and the productive sector, as well as environmental management agencies.

The DNA barcode of Life Initiative, since its inception in 2003, stated as an effective method to uncover biodiversity in the life realm, including for macroalgae. Collected data proved useful for uncovering cryptic species, assessing the relationships with neighbouring floras, monitoring allochthonous introductions, and for the identification of problematic taxa to boost further taxonomic research. DNA barcode identifications contributed to new records to DNA libraries of life, such as the BOLD catalogue, with associated metadata freely available to the scientific community and the general public. Furthermore, in the context of environment management, the added value of DNA barcode-assisted identifications is that they give objective and verifiable data, and each specimen is unequivocally linked to a permanent genetic label regardless of any subsequent taxonomic or nomenclature variation, an essential strength if we consider that floristic lists are extensively used by ecologists and environment agencies as the basis for monitoring studies.

# Patterns and mechanisms of plant diversification driven by chromosome shifts in Mediterranean lineages

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The Mediterranean Basin region, with 25,000 plant species, is a worldwide hotspot of biodiversity. Despite the questionable important role of chromosome transitions in plant evolution and diversification, we do not still have an understanding of the possible relationship between chromosome evolution and lineage diversification in the Mediterranean Basin.

We have studied here patterns of diversification, chromosome number transition (either polyploidy or dysploidy) and the relationship between the two and also with other important traits for three angiosperm lineages with the highest species richness in the Mediterranean Basin: *Linum (Linaceae)* and *Centaurium (Gentianaceae)* at macroevolutionary level, and *Carex gr. laevigata (Cyperaceae)* at microevolutionary level. Specifically, at macroevolutionry level, we have modeled on the phylogenies (based on several DNA regions or thousands of RADseq loci) chromosome evolution (ChomEvol model), and jointly with diversification rates (ChromoSSE model) and with important traits (ChromePlus model) to test several traditional hypotheses onf chromosomal evolution. At microevolutionary level, we have conducted a landscape genomic analysis (redundacy analysis or RDA) that test the association of genome loci with environmental variables.

We found a mixed pattern at macroevolutionary level. Whereas cladogenetic process are not linked to chromosomal transitions in *Linum*, in *Centaurium*, polyploid transitions are one of the major drivers of diversification. In *Linum*, important traits such as habit and biogeography (but not reproductive systems) are dependent on chromosome evolution. In *Centaurium*, heteroploid (allopolyploidy) and homoploid hybridation are crucial evolutionary processes to understand its diversification process. At microevolutionary level, we have found that Pleistocene glaciations explains, at least partially, the distribution of the karyotypes in *Carex gr. laevigata*. We have also found significant association of loci across the genome and karyotypes with environmental variables related to temperature and precipitation that suggest adaptive chromosomal evolution.

New chromosomal variants could have been key for the successful establishment of plant lineages during the Mediterranean onset and the posterior Quaternary climatic oscillations. The biodiversity of the Mediterranean Basin is partly influenced by the chromosomal diversity of its lineages.

# Taxonomic and evolutionary studies on Mediterranean plant groups in Greece: the examples of *Centaurea* (*Asteraceae*) and *Limonium* (*Plumbaginaceae*)

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Identifying and classifying the diversity of plant lineages in the Mediterranean region, and exploring their evolutionary history, contribute to the understanding of how and why this extraordinary diversity has accumulated in one of the world's biodiversity hotspots. Focusing on the rich flora of Greece, we use *Centaurea* L. (*Asteraceae*) and *Limonium* Mill. (*Plumbaginaceae*) as examples of genera with remarkable taxonomic complexity for which we employ state-of-the-art tools and methods to explore species limits and relationships and produce species descriptions and identification keys.

For *Centaurea*, which is the most species-rich genus in Greece, we integrate morphological and molecular data to delimitate taxonomic units, identify their relationships, and provide a revised taxonomy. We develop an exemplar workflow for integrative taxonomy and biodiversity data management, using the virtual research environment of the Platform for Cybertaxonomy, which covers all aspects of the taxonomic workflow (i.e., taxonomic data processing and storage, specimen-level recording and storage of character data, generation of taxon characterizations, identification keys, distribution maps etc.). Hybrid capture with high-throughput sequencing (Hyb-Seq) is employed to generate molecular data for phylogenetic inference. Combining newly generated data from morphological and phylogenomic analyses with other available data (e.g., chromosome numbers, geographical distributions), we will produce an up-to-date treatment of *Centaurea* for the printed and online versions of the Flora of Greece.

For *Limonium*, we focus on the endemics of Crete and adjacent islands (Southern Aegean Island arc) that represent a complex group of species characterized by polyploidy and asexual reproduction via apomixis. We aim at testing the monophyly and limits of the currently described species, clarify the evolutionary origins of these lineages, and disentangle their relationships. We used newly assembled transcriptomes of *Limonium* to identify low copy nuclear loci and generate molecular data with the target capture method for 31 apomictic polyploid Aegean endemics, few diploid and triploid Mediterranean species, and two outgroup species. Our analyses revealed that the Aegean endemics are of hybrid origin (allopolyploids) with two or three parental subgenomes. Their reticulate evolution is represented in a phylogenetic network showing that the Aegean apomicts originated by a single or multiple hybridization events. Most of the Aegean species were monophyletic, while few changes in taxonomic circumscriptions are proposed following morphological observations and phylogenomic results.

### Historical introgression and evolutionary parallelisms drove diversification and trait evolution in the European endemic genus *Soldanella* (*Primulaceae*)

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Historical introgression is a prominent mechanism driving lineage diversification and has considerable implications for the evolution of intrinsic and extrinsic traits. Introgression-induced reticulations often lead to incongruent trait patterns between gene trees and species trees, giving rise to the erroneous perception of homoplasy. In this study, employing organellar and nuclear genomic data in combination with cytogenetic approach, we investigated the impact of historical introgression on the diversification and trait evolution within the genus *Soldanella (Primulaceae)*. To analyse trait evolution, we employed a comparative method for phylogenetic networks, in conjunction with inferred patterns of historical introgression within the genus. Our findings reveal extensive historical introgression, with dwarf species predominantly associated with snowbed communities, Carpathian species exhibiting broad ecological adaptation, and species from Mediterranean mountains in the eastern and southern Balkans being significantly affected. Notably, Balkan species (*S. pindicola, S. chrysosticta*, and *S. rhodopaea*) can be regarded as hybridogenous entities.

Using network-based ancestral state reconstruction, we demonstrate that dwarfic singleflowering phenotypes, dysploid cytotypes, and ecological generalisms have independently evolved at least twice. The latter originated in the most recent common ancestor of the genus and served as a preadaptation, facilitating subsequent activation of specific adaptations to forest or alpine zone habitats in response to climatic shifts, thus promoting adaptability to new environments. The recurring occurrence of stoloniferous growth in hygrophilous highmountain species remains uncertain, necessitating further investigation to discern between adaptive introgression and parallel evolution.

Phylogenetic regression analyses suggest that introgression events between taxa, occurring within specific environments at the margins of their distribution ranges (Mediterranean mountain ranges), may contribute to genome size expansion. Our study underscores the complexity of trait evolution reconstruction, highlighting the interplay among various evolutionary phenomena. Hence, it is crucial to consider all these factors to disentangle the evolutionary history of traits.

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# Phylogenomics and the biogeography of the Old-World Xanthium strumarium (Asteraceae)

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*Xanthium* L. is a wind-pollinated genus of subtribe Ambrosiinae (Asteraceae). Together with a few other genera of the subtribe (e.g., *Ambrosia* L., *Iva* L.), it is characteristic for having spiny, female capitula (burs), dispersed by animal or water streams. Some lineages of the genus have a pronounced ruderal character and have become weeds with an important economical impact in some part of the world. Burs can be found as contaminant in raw fibres (e.g., cotton, wool), and grain loads. Due to this capability, and the last few centuries intensification of human trade, several lineages of the genus have nowadays a cosmopolitan distribution.

However, *Xanthium strumarium* L. is the only species of the genus native to the Old World. Indeed, it is one of the very few species of the whole subtribe Ambrosiinae with a non-American origin. Until recently, *X. strumarium* was distributed in the Mediterranean, in Central Europe, in Ethiopia, in the Middle East and in Asia up to Japan. It is unclear, how and in which time frame the ancestor of the species reached the Old World, and how it spread throughout the continent.

In this study, we used target enrichment of hundreds of nuclear loci and complete plastome sequences, along with phylogenetic reconstructions and biogeographical analyses, to reconstruct the origin and the routes followed by *X. strumarium* during its colonization the Old World. We sampled about 40 herbarium specimens, collected in the last two century across the entire distribution range of the species.

The origin of *X. strumarium* must be recent, as testify by the relatively low genetic variation found in the plastomes and in the nuclear regions. This is particularly evident if compared with the variability found in the American species of the same genus. However, it is still possible to recognise in the species a certain geographic structure and pattern of distribution. A more recent event of (human-mediated) backword colonization of the species in South America (south-eastern Brazil) was corroborated by the results of our analyses.

## Lumping and splitting: how an integrative approach can guide taxonomists in species delimitation

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Among plant taxonomists, taxa delimitation is surely one of the most debated issues. While lumpers tend to recognize fewer more inclusive taxa, splitters recognize a higher number of less inclusive taxa. This diversity of perspectives generates problems that affect not only taxonomy, but also related disciplines. For instance, since species serve as the fundamental unit for conservation projects, an unstable taxonomy can greatly impact the effectiveness of conservation efforts.

To overcome this problem, several authors started adopting an integrative taxonomic approach, in which species are considered as hypotheses to be rigorously tested by adopting multiple independent lines of evidence. This allows for a more reliable taxonomy at species level, by considering multiple biological aspects. For instance, morphometric analyses provide insights into morphological variation, whereas cytogenetic and molecular approaches can unveil evolutionary relationships among units of diversity. Additionally, niche studies can allow evaluating the degree of ecological differentiation.

Integrative taxonomy was recently applied to three critical species groups in the *Santolina chamaecyparissus* L. complex (*Anthemideae, Asteraceae*), endemic to the western Mediterranean. The first case concerns the tetraploid and hexaploid populations of *Santolina* in Corsica and Sardinia, traditionally recognized as two distinct species. By adopting morphometrics, seed morpho-colorimetrics, molecular systematics (plastid markers: *trnHpsbA, trnL-trnF, trnQ-rps16, rps15-ycf1, psbM-trnD,* and *trnS-trnG*), and climatic niche analysis, it was concluded that these two species cannot be separated. The second case concerns *S. benthamiana* Jord. & Fourr., a diploid species growing on both sides of eastern Pyrenees (France and Spain). By using a similar approach, it was highlighted that taxonomically doubtful populations growing on average at lower elevations are more related to *S. decumbens* Mill. Accordingly, these populations were recognized as a distinct species, namely *S. intricata* Jord. & Fourr. The last case concerns *S. decumbens*, another diploid endemic to Provence (southern France). In this species, three allopatric morphotypes were detected and recognized as distinct subspecies based on slight ecological and molecular differentiation.

Accordingly, it was possible to elaborate more reliable taxonomic hypotheses concerning species and subspecies within the *Santolina chamaecyparissus* complex.

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# Intra- and inter-population phytochemical variability of *Teucrium marum* (*Lamiaceae*) in Sardinia: relevant for chemotaxonomy or just determined by ecological factors?

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Secondary metabolites produced by vascular plants, like terpenoid and phenolic compounds, may vary among and within populations, generating distinct chemical phenotypes, commonly called chemotypes. Whether the chemical composition of secondary metabolites has a taxonomic significance (chemotaxonomy) or is just determined by local/regional ecological factors, is a key question intriguing botanists. Lamiaceae are of particular interest to evaluate whether chemotypes have a taxonomical relevance or are determined by ecological factors, since many species are polymorphic in their essential oil compositions.

Here we investigated intra- and inter-population phytochemical variability of the western Mediterranean endemic *Teucrium marum* L., a dwarf aromatic plant well-known from the phytochemical point of view, but insufficiently investigated regarding the ecological and/or chemo-taxonomic factors that influence the chemical diversity of its essential oil composition. The specific goal was to evaluate whether geographical distribution, elevation, climatic factors and/or the soil substrate determined the phytochemical variability of *T. marum* along a gradient from coastal to mountain wild populations on the island of Sardinia (Italy).

During their flowering period, fresh *T. marum* aerial parts were randomly collected from ten individuals in six different localities in Sardinia. Dried leaf samples were hydrodistillated using a classical Clevenger apparatus to obtain the corresponding essential oils. The composition of each essential oil was chemically characterised by gas chromatography coupled to mass spectrometry. In total, ninety compounds were identified. Statistical analyses showed significant differences in phytochemical essential oil composition among and within the six studied populations. The two main compounds of the essential oils were DOLICHODIAL and (*E*)- $\beta$ -CARYOPHYLLENE, which followed opposite patterns, the first compound being much more abundant in coastal populations (Alghero, Cagliari, and Costa Paradiso), and the second in mountainous ones (Osilo, Limbara, and Urzulei).

Taken as a whole, the results presented here showed that environmental and geographical conditions determined the chemical variability of essential oils in *T. marum*, emphasizing a clear grouping of coastal vs. mountainous populations. However, no morphological variability among coastal vs. mountainous populations was detected, and therefore further studies will be necessary to assess the chemotaxonomic significance of our findings. Nevertheless, here for the first time, it was shown that the essential oil composition of Sardinian *T. marum* was unique among other Mediterranean known compositions for this species, particularly for the high level of DOLICHODIAL in coastal populations. Furthermore, this work contributed to reveal chemotype distribution of this species in Sardinia.

# Towards defining the genetic variation of *Limonium cuspidatum* (*Plumbaginaceae*), a protected taxon in SouthFrance, using microsatellite markers

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Limonium cuspidatum (Delort) Erben shows a restricted distribution range, from Narbonne to Marseille in southern France, where it is protected at national scale. Morphology alone is not sufficient to clearly distinguish this taxon from three others species cited from the same environments in southern France: L. confusum (Godr. & Gren.) Fourr., L. legrandii (Gaut. & Timb.-Lagr.) Erben, L. densissimum (Pignatti) Pignatti, all belonging to the L. confusum aggregate. Here, we aimed to define the genetic variation of L. cuspi*datum* and to clarify its genetic relationships to the other three species of aggregate. We carried out a first genetic screening of 31 populations (23 from France and 8 from Spain), using 6 Simple Sequence Repeat (SSR) loci. We identified three genetic groups. A first group includes Iberian-Provençal population, from the salty mud of the Bouches-du-Rhône (France) to the Tarragona and Ebro Delta region (Spain). The second genetic group includes populations of the salt marshes located in the Languedoc coast between Leucate and Vendres (France). The third genetic group includes three disjunct populations, which however share similar ecological conditions. Our results enable us to identify homogeneous genetic groups that will serve as accessions for a phylogenetic study based on extensive sampling of the chloroplast genome.

#### More than 20 years of flora conservation in Menorca: landscape changes and management

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Flora conservation projects have been carried out in Menorca for more than 20 years. During this time, the conservation status of dozens of plant taxa, threatened in the island territory, has been monitored and assessed. All this means that the volume of accumulated information is important, in many aspects: chorology, censuses, threats, companion species, etc.

Possibly one of the most interesting aspects is how can be appreciated the changes in the distribution of species and their behaviour. In Menorca, the abandonment of agricultural and livestock activity is causing an increase in forest areas and a transformation of the vegetation landscape. One consequence is the redistribution of habitats and of certain plant species. Most of these changes are positive for endemic and threatened species, although there are also a few cases with a negative trend.

Men arrived in Menorca about 4,000 years ago. The only information we have about the pre-human plant landscape is what can be inferred from the chronological pollen paleo sequences. In the different studies of this type that have been carried out, there are coincidences in a change in vegetation with the arrival of men. Woody species decrease, mainly arborescent, and herbaceous and pioneer species increase.

The absence of a megafauna that could alter the forest masses suggests that pre-human Menorca was largely covered with forest vegetation, whether lighter or thicker. The afforestation process that exists today on the island could be like a return to that pristine island.

In these 20 years it has been observed that some endemics have increased the extent of their populations: *Cyclamen balearicum*, *Paeonia cambessedesii*, *Teucrium asiaticum*, etc. Others have changed their habitat, for example, from strictly rupicolous to terrestrial: *Hippocrepis balearica*, Lomelosia cretica, *Teucrium asiaticum*. At least one endemic and threatened species, *Malva minoricensis*, shows a negative trend in part of the island.

At the same time, in these 20 years in Menorca there have been changes in the socio-economic field: consolidation of tourism as the main economic income, increase in the human population, increase in diffuse pressure, etc., which have effects on the flora threatened: more ways for the penetration of invasive plants, alteration of sensitive habitats, expansion of ruderal vegetation, etc.

All this information and observations make it possible to reach conclusions, that can be used to propose new management strategies, which are more solid, coherent, and better for long-term conservation.

# Facilitated adaptation as a conservation tool for plant adaptation to climate change

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Adaptive responses at the genetic level may be the only way for certain species or populations to survive the environmental conditions imposed by climate change. However, given the speed at which environmental changes are occurring, these responses may not be sufficient to ensure the survival of certain plant species in their current habitats. It is therefore necessary to look for new tools to help us to conserve biodiversity. One of those tools is assisted migration, which involves the deliberated movement of individuals to an area outside their current range, where future environmental conditions are predicted to be favourable. As this type of action can involve many ecological risks, other strategies have been proposed to promote the *in situ* adaptation of organisms, such as artificial selection and assisted gene flow. These two strategies fall under the concept of facilitated adaptation and aim to increase the adaptive potential (the ability of organisms to evolve and adapt to new environmental conditions) of populations, in response to climate change. The concepts underlying facilitated adaptation approaches, such as artificial selection and gene flow, are not new. However, their direct application in conservation is still in its infancy, probably due to concerns about their use. We used the legume Lupinus angustifolius L. as a study species to provide a proof of concept for these novel strategies. We have conducted artificial selection and assisted gene flow experiments under both controlled and natural conditions and evaluated the effects of these techniques at both phenotypic and genotypic levels. Our experiments showed that the use of assisted evolutionary techniques using artificial selection and assisted gene flow is promising in theory, but complex to implement due to the polygenic nature of most traits, the different trade-offs and correlations between traits, and genotype-environment ( $G \ge E$ ) interactions. However, with the right knowledge and tools, this efficiency could be improved and limitations overcome.

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#### Design and development of genetic reserves for plant conservation

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Current approaches to biodiversity conservation are largely based on ecosystems, ecological communities, and species, with less attention on genetic diversity and the evolutionary continuum from populations to species. The design of genetic reserves networks for plant conservation aims at conserving not only the species-level diversity, but also the genetic diversity at the intraspecific level, contained amongst plant populations. In the context of climate change, it is essential to provide insight into the adaptive genetic diversity present in plant populations and make it available to be used in a wide range of potential conservation actions and other applications (e.g., reinforcements, reintroductions, translocations, facilitated adaptation, plant breeding...). Prior to the design of the network, a clear definition of the target plant species and how their populations are distributed across the selected territory is required. Subsequently, the adaptive genetic component must be associated with each population by considering environmental pressures in their inhabiting sites. Assuming that the patterns of adaptive genetic diversity are mediated by the heterogeneity of environmental pressures across the territory, we can estimate the distribution of adaptive genetic diversity amongst populations through the generation of environmental maps built from abiotic variables. Once both the targeted species and environmental proxies of adaptive genetic diversity are defined, different criteria for the selection of sites to establish genetic reserves can be applied. These criteria involve a) efficiency, both in covering the highest number of species and potential genetic diversity in the lowest number of places, but also in the use of available human and economic resources; b) taking into account idiosyncratic and legal aspects related to the management of sites or species and c) mid or long-term viability of the proposed reserves. We show some examples involving the design of genetic reserve networks of crop wild relatives and discuss the potential benefits as well as the limitations of this approach.

# Celebrating a centennial – 100 years of struggle for the preservation and rehabilitation of plants in Israel

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Preservation of nature in Israel is now days in the hands of a well-established authority (INP), activated by law and associated with international nature preservation enteritis. This however was not so to begin with. It took many decades to amalgamate civil initiatives and sporadic governmental regulations into a national endeavor.

1922 - first preservation act under British mandate, assigning Umm-Safa Forest in Samaria a "Forest reserve". This however was not a mere act of preservation since forest reserves were declared to serve as a hunting area for British civil servants.

1924 - the "Hunting Order" was published, regulating, and restricting hunting.

1926 – the "Trees Order" forbade cutting protected trees, mostly larger wild species, without permission by the forests officer. It was issued recognizing the fast-dwindling natural resources under heavy utilization and rapid modern development.

Yet in 1926 Alexander Eig called for preservation before it is too late: "Those concerned about our land's nature and its future, should join forces to create a nature preservation society whose main objective is to stand guard for the vegetation of Palestine".

Eig was then a traveling coach teaching plants recognition and basic botany to teachers. In few years he obtained his Ph.D. and joined the Hebrew University faculty where in 1931 he founded the botanic garden, dedicated to the natural plant associations of Palestine. Eig died in 1939, but his teachings live on.

- 1933 Heinrich Mendelssohn who immigrated from Berlin to study zoology became the most eminent preservation activist of Israel until his death in 2002. He inspired many to strive for nature protection.
- 1948 establishing of the state of Israel.
- 1953 Israel's Society for Protection of Nature (SPNI) established by Mendelssohn and two colleges – Amotz Dafni and Azaria Alon. It was a very effective NGO, numbering over 120,000 members, and leading many successful campaigns.
- 1955 first reservation law, "Wildlife protection law".
- 1964 "Israel's Nature Preservation Authority" established, finally an enforcing authority.
- 1986 a group of amateurs and students launched a "Rare Plants Survey" which soon became a national project.

Hence it is in the hands of NPA, and efforts continue up to date with an adaptable "Red list", many refuge gardens, and rehabilitation projects.

### Populations genomic analysis of *Primula palinuri (Primulaceae)*, a narrow endemic Mediterranean plant species on the edge of precipice

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Primula palinuri Petagna is a relict species endemic to Italy, whose sparse populations can be found along the vertical cliffs overlooking the sea along the Tyrrhenian coast for among eighty kilometres. Habitat peculiarity defines P. palinuri as the only Italian Primula adapted to live in a coastal environment. Its ecological refuge has been so advantageous in successfully overcoming the challenges of time, also by exploiting evolutionary strategies such as polyploidisation, but today it makes the species at risk of extinction due to tourismrelated anthropic activities in the area. The aims of this study were (i) to reconfirm the known population of *P. palinuri* along the Tyrrhenian coast, (ii) to record new populations if any, (iii) to define any phenotypic plasticity, (iv) and to carry out a population analysis of the species throughout its distribution area to determine intraspecific genetic variation. For these purposes, survey and sampling campaigns were conducted among the stretch of coastline concerned. Known populations were reconfirmed and an unreported population was found along the coast for a total of 8 populations identified and sampled. After proper authorization, the sampling phase of the plant material was conducted using a rigorous methodology aimed at minimising damage to the organs, taking the minimum portion of leaf tissue required for DNA extraction (approx. 0.2 g), without disrupting population stability. Genetic diversity, population genetic structure and gene flow of P. palinuri populations were evaluated by Whole-Genome Sequencing (WGS) which provides more information than the reduced representation methods. The 8 populations identified and sampled, consisting of an estimated 30 to 200 individuals each. The populations were extremely fragmented and spatially divided by large geographical barriers represented by the structure of the coastline and, in the case of the Dino Island population, by the sea. Phenotypic plasticity of floral characters was identified among the analyzed populations, hypothetically attributable to adaptations due to the selective pressure caused by the scarcity of pollinators and mates. Prominent among these adaptations is heterostyly, already highlighted in previous works. Data obtained by population genomic analysis allowed the definition of population structure and the related genetic flux. It has showed that P. palinuri identified and sampled populations can be divided into defined and well clustered groups. Our population genomic study tackles a central issue in evolution and conservation biology and we expect that it will be useful to help safeguard the remaining genetic diversity reported for this unique genetic resource.

Our thanks to the Park Authority of Parco Nazionale del Cilento, Vallo di Diano e Alburni and the Reparto Carabinieri Parco di Vallo della Lucania for their trust, availability and support during the survey and sampling operations.

# Use of micropropagation techniques for the conservation of endangered species with reproductive problems

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Conservation of endangered, rare, and endemic plant species faces multiple threats including habitat loss, overexploitation, invasive species, nitrogen deposition, and climate change. Endemic taxa, with limited geographical ranges, are particularly at risk of extinction. *In situ* conservation, which aims to protect and preserve populations within their natural habitats, is the most effective approach. However, it may not always be feasible due to various constraints. In such cases, *ex situ* techniques are adopted as a complementary measure to ensure the survival of threatened species. Micropropagation protocols play a crucial role in *ex situ* conservation programs by enabling the propagation of endangered plants. This approach offers advantages such as year-round availability, rapid propagation, and production of disease-free plants. Our research focused on developing efficient micropropagation protocols for *Adenostyles* Cass., *Hieracium* L. (*Asteraceae*) and *Erica* L. (*Ericaceae*). Specifically, we developed a micropropagation protocol for *A. alpina* subsp. *macrocephala* (Porta) Dillenb. & Kadereit, endemic to Calabria, *H. lucidum* Guss., and *E. sicula* Guss. subsp. *sicula*, both punctual endemics to carbonate cliffs of NW Sicily.

The impact of different plant growth regulators on leaf explants was evaluated. For callus induction in *Adenostyles* and *Hieracium*, we tested various concentrations of 1-Naphthaleneacetic acid (NAA) and 2,4-Dichlorophenoxyacetic acid (2,4-D) in combination with 6-Benzylaminopurine (BAP) and/or meta-Topolin (mT). For *Erica*, BAP, mT and Thidiazuron (TDZ) were tested in combination with NAA and 2,4-D. Root induction was assessed using media supplemented with Indole-3-butyric acid (IBA) and NAA. High callus induction and increased shoot regeneration were obtained for *A. alpina* subsp. *macrocephala* using Murashige and Skoog (MS) medium supplemented with 3 mg L<sup>-1</sup> BAP and 1 mg L<sup>-1</sup> NAA. Effective root induction was achieved using hormone-free MS medium and IBA at varying concentrations.

Before implementing any reintroduction program, it is essential to evaluate the genetic diversity of the regenerated plants due to somaclonal variations during in vitro culture. When these variations cover the original population's diversity, they can be useful for maintaining genetic variability in the reinforced population. The findings of this study have important implications for the development of micropropagation protocols not only for the taxa investigated, but also for others specialized to Mediterranean conditions. Micropropagation techniques provide a means to collect and preserve endangered plants, thereby reducing the risk of extinction and supporting conservation efforts. By utilizing these techniques, we can contribute to the conservation of threatened species and ensure the long-term survival of their genetic diversity.

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### Origin and Evolution of Herbaria in the 16<sup>th</sup> Century

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The origin of herbaria represented a revolution in the history of botany, and more in general in the history of natural science. Indeed, herbaria are the key instrument and the very icon of scientific botany.

Where and when this scientific revolution began, is an open question, since many of the early herbaria were lost or dispersed through time. Nevertheless, convincing evidence points to Bologna and/or Ferrara as the cradle of this innovation. Ferrara, the seat of the Court of Este, was a hotspot of scientific humanism during the first half of the 16<sup>th</sup> century. In this context, some evidence indicates that the physician and humanist Nicolò Leoniceno, a forerunner of modern science who taught in Ferrara for sixty years, played a role in opening the way to this innovation. Beside Ferrara, Bologna was a center of diffusion of the *ars herbaria*, and Luca Ghini an undisputed leader.

From the primary center, the art of making herbaria spread rapidly in Italy and then throughout Europe: by 1560 at least twelve major herbaria (nine of them in Italy) had been accomplished or incepted; by the end of the century, we have notice of about 20 herbaria (14 of them in Italy), either preserved or dispersed.

The primary scope of herbaria was to anchor each plant name to a plant specimen. Reference to a permanent specimen was, indeed, the only way to overcome the ambiguity inherent to verbal descriptions or even to illustrations. In the earliest herbaria plants were labeled with the names given by Dioscorides, considered by definition as the "correct" ones.

Starting from the half of the 16<sup>th</sup> century, however, a rich editorial activity arose, with the production of major works that described all species known hitherto, along with several new ones, adopting different nomenclatures. This circumstance induced some authors to label their herbarium specimens with a more or less rich synonymy. Such trend proceeded with time, so that, at the end of the century, some herbaria listed long lists of synonyms for each species.

Last but not least, herbaria offered an unprecedented opportunity: plant specimens associated with their names could be sent everywhere in Europe, so that scholars in different countries could know exactly what meaning their colleagues abroad gave to any plant name: so, for the first time in history, herbaria allowed to establish a common language, that was the prerequisite for the inception of a European scientific community.

### How to approach the unknown? Elucidating the origins of the 16th-century En Tibi and Rauwolf book herbaria

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Historic herbaria often have imperfectly known origins. Finding out who made them, when, where (and for whom) is a fascinating process that requires an integrative mindset. Here we present methodological approaches to assess the origins of two 16<sup>th</sup>-century plant collections from the Mediterranean region: the En Tibi and Rauwolf book herbaria. These two collections, comprising in total five book volumes, became property of Leiden University in 1690, and are currently preserved in the herbarium of Naturalis Biodiversity Center in Leiden, The Netherlands.

The En Tibi herbarium is a large luxurious leather-bound book with golden details that contains about 500 dried plants. This enigmatic collection was suggested to have been made in Ferrara in 1542-3. Its maker was long unknown. Our interdisciplinary approach that combined plant taxonomy, the study of paper, watermarks and book binding, history, paleography, Neolatin literature and forensics showed that this precious collection was actually made in the area of Bologna around 1558. It also revealed that many people were involved in the making process of the book which was intended as a gift to a person in power, possibly the Habsburg emperor, Ferdinand I. The plant material was provided by the physician and botanist Francesco Petrollini.

The Rauwolf herbarium comprises four book volumes with about 800 dried plants. It was compiled by the German physician, botanist and traveller Leonhard Rauwolf and later purchased by the Habsburg emperor Rudolf II. Here we focus on the first three book volumes, which Rauwolf compiled in 1560-3 in southern France and northern Italy during and right after his medical studies at the University of Montpellier. A study of the paper watermarks and handwritten texts accompanying the plant specimens, helped us reconstruct the gradual process that Rauwolf followed to compile the three books in France, Italy and later in Germany. A closer look at the different handwritings revealed that the French volumes were actually largely compiled by Rauwolf's classmate and field companion, Johan Bauhin. This finding changed our perception of how 16<sup>th</sup>-century herbaria were made.

We suggest that a multidisciplinary methodological approach, involving experts in disciplines of both science and humanities, is necessary in order to elucidate the origins of anonymous historic herbaria. Many historical collections are being digitized and become available online. Their increasing accessibility greatly facilitates research efforts to elucidate the contents and origins of these treasures of our scientific and cultural heritage.

# Taxonomy in the spotlight: integrating herbarium-based research and genomics to monograph the world's plants.

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Throughout history, taxonomy has contributed crucially to expanding our understanding of nature. By studying and organising the diversity of life on Earth, taxonomy has helped shape our comprehension of the natural world: it has shed light on the relationships between species and with the environment and has provided the necessary evidence to support scientific theories and concepts. The advent of new technologies, together with changes in publication trends, has however changed the focus of biodiversity studies from an integrative discipline such as taxonomy to other highly technological, often model-based disciplines (genomics, distribution modelling, etc.). This, in turn, has led to a progressive detachment between taxonomy and the rest of scientific disciplines studying biodiversity, with taxonomy often contemptuously regarded as a mere practice of "stamp collecting". This detachment between taxonomy and other research disciplines has important, often overlooked consequences: most plant groups, especially in the tropics, have never been comprehensively studied, and the world's herbaria accumulate a large number of misidentified specimens. These specimens are the main source of data, for example, for global conservation studies based on data obtained through databases such as GBIF, which likely also contain a significant number of misidentified records.

We need to bring taxonomy back to the spotlight. In this talk, I will argue that we need faster, more comprehensive taxonomic studies, and we need to integrate herbarium-based morphological studies with molecular data throughout the taxonomic decision process. To illustrate this, I will summarise the key findings of a monographic study of the megadiverse genus *Ipomoea* L., which involved the analysis of approximately 25,000 herbarium collections. I will present a pipeline, developed as part of our work, that integrates specimen based morphological studies and techniques of phylogenetic and genomic analysis to develop more robust species delimitation hypotheses and accelerate biodiversity studies.

Beyond taxonomy, the results of a monographic study have implications for other disciplines such as archaeology or food security. I will specifically show how this integrated approach using herbarium specimens enabled several important discoveries on the origin and evolution of sweet potato (*Ipomoea batatas* (L.) Lam.), one of the most important food crops worldwide.

## Herbaria for the next generations, the exemple of Litardière herbarium and herbonautes in Maghreb

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In terms of conservation, the first question is what we know about about species ecology and distribution, but identificaction is also necessary. In North Africa, an OpenAccess website (efloramaghreb.org) is now available for acquisition and georeferencing herbarium samples (and observations), for sharing information. It contains around 100'000 herbarium samples from different international institutions. This tool is usefull to assess the state of species knowledge and identify those to be researched, or even to define areas to be prospected. Herbaria are an essential source of information, as taxonomy and nomenclature can be verified and updated. The availability of these collections for future generations is also essential.

Our questions concerns how to acquire naturalist observations, particularly for flora, using the new participative tools such as inaturalist, which are taking off in leaps and bounds and enabling everyone to get involved, discover and finally learn to protect biodiversity. These approaches are undoubtedly complementary, as photography allows many and quick georeferenced observations to be made, and is very useful for identifying some taxonomic groups (e.g. *Amarantaceae* or *Orchidaceae*, which are very difficult to preserve in herbaria), but is almost impossible for others, such as *Festuca, Erodium* and many other genus. At present, the number of participants and observations in the Maghreb is several thousand times greater than the number of naturalists making herbarium collections. What will be the futur of the data acquired and its validation in a few years' time?

In order to evaluate these different sources of information, we will take the example of the collections of the Litardière herbarium on the Maghreb, created 100 years ago, with the genus *Festuca* which is not atractiv; and the collections of *Erodium* inventoried by Herbonauts from the MNHN, a complex genus needing akenes magniying observation but much more attractive photographically; then we will compare the relevance of these identifications and distibutions in relation to naturalist observations or certain aggregated data available on the Web.

Litardière's herbarium was deposited in Geneva in 1986, and his North African collection contains 7,000 specimens, corresponding at the time to 3'822 taxa. The data aquisition process made it possible to update the nomenclature of the collection and identify types, resulting in a list of 3050 taxa. This shows the need to be able to re-examine and revalidate the observations.

Our conclusions concern the efforts that need to be made to gain a better understanding of the ecology of species, the various tools available, and the efforts, resources and collaboration required to achieve this.

# Forest wildflowers bloom earlier as Europe warms: lessons from herbaria and spatial modelling

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Today plants often flower earlier due to climate warming. Herbarium specimens are excellent witnesses of such long-term changes. However, the magnitude of phenological shifts may vary geographically, and the data are often clustered. Therefore, large-scale analyses of herbarium data are prone to pseudoreplication and geographical biases. We studied over 6000 herbarium specimens of 20 spring-flowering forest understory herbs from Europe to understand how their phenology had changed during the last century. We estimated phenology trends with or without taking spatial autocorrelation into account. On average plants now flowered over 6 days earlier than at the beginning of the last century. These changes were strongly associated with warmer spring temperatures. Flowering time advanced 3.6 days per 1°C warming. Spatial modelling showed that, in some parts of Europe, plants flowered earlier or later than expected. Without accounting for this, the estimates of phenological shifts were biased and model fits were poor. Our study indicates that forest wildflowers in Europe strongly advanced their phenology in response to climate change. However, these phenological shifts differ geographically. This shows that it is crucial to combine the analysis of herbarium data with spatial modelling when testing for long-term phenology trends across large spatial scales.

# Digitization of historical herbaria. A case study from the Herbarium of Michele Guadagno at Pisa (PI)

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The herbarium digitization process - i.e. the capture of images and metadata from specimen labels - is an essential first step in transforming this vast amount of data associated with a physical specimen into flexible digital data formats accessible, usable, and useful since it allows information to be summarised, categorized and manipulated, in order to retrieve specific information, otherwise hidden in an overwhelming pile of specimens. The digitization of herbarium specimens is of primary importance in studies of different biological disciplines like taxonomy, biogeography, nature conservation, biographies of botanists and many others. In this framework, the Herbarium of the University of Pisa (international acronym PI), at the end of 2018 started a process of digitization focusing on one of its most relevant collection: the herbarium of Michele Guadagno (1878–1930), bought in 1939 under the direction of Alberto Chiarugi (1901-1960). Michele Guadagno was born in Naples (Campania, southern Italy) and showed from an early age a special attitude for botany. He studied flora and vegetation of different areas of southern Italy, building a large herbarium including specimens collected by himself mainly in central and southern Italy, plus many specimens obtained through exchanges with Italian and foreign botanists. The herbarium is composed by 547 packages of vascular plants, of which 508 organized according to a systematic criterion, and other 39 organized according to collection localities or correspondents. Metadata were entered into the online database Virtual Herbaria JACQ (http://www.jacq.org/), and mirrored into a personalized virtual herbarium of the Botanic Museum (https://erbario.unipi.it/erbario/index). The digitization process allowed to extract and summarize information about collectors, localities, collection dates, and taxonomic information about the specimens, including nomenclatural types. After the completion of the digitization process, the number of sheets preserved in the Herbarium amounts to 44,345. Besides Guadagno, who collected 42% of the specimens, other 1,102 collectors are represented. Most of the specimens were collected in Europe (91%), but all the continents are represented. Italy is the most represented country (59%), followed by France, Spain, Germany, and Greece. All the digitized specimens cover a time span of 99 years, from 1830 (48 years before the birth of Guadagno) to 1929, whereas the specimens collected by Guadagno range between 1889 and 1928. Furthermore, we traced 134 herbarium sheets associated with documents, among which 75 drawings handmade by Guadagno, 34 letters from various corresponding authors, 16 copies of publications, and 14 copies of published iconographies.

#### Herbarium specimens in the web of scientific data in 2030, a forward look

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Worldwide there are 3100 herbaria registered in Index Herbarium from which about one third is housed in Europe. Finding out what material is present in all these herbaria is a time consuming task today, but DiSSCo, the Distributed System of Scientific Collections is developing ELViS, the European Loans and Visits System to act as a one-stop shop for access to the herbaria and other natural history collections in Europe in the future. A first version of ELViS has already been used in the EU-funded SYNTHESYS+ project to provide transnational access to collections from 22 institutions and over 1500 access requests were made in two calls organised by the project.

While access to the physical objects is often necessary, for many scientific use cases access to digitised data is sufficient and more efficient. Digitisation in herbarium collections has been progressed faster than in other natural history collections as the 2D nature of herbarium sheets makes large scale digitisation using conveyor belts relatively straightforward and a complete digitization of herbaria seems achievable. However ,surveys learn that still a lot remains to be done. While the biggest natural history museums in Europe have digitised about 50% of their botanical specimens, for example in Germany only 13% of the herbarium specimens was digitized in 2019 when taking also the smaller collections into account.

The current rapid developments in Artificial Intelligence and robotics provide new opportunities to further industrialise digitisation and lower the costs, which helps to speed up scientific discoveries. DiSSCo is implementing a FAIR Digital Object infrastructure for Digital Specimens that will be Fully AI Ready and is planned to be in operation by the start of the infrastructure in 2025. The data infrastructure will provide for machine and expert annotations to aid digitisation and to enrich the specimen data by (re-)connecting derived and related data to the Digital Specimen. The infrastructure aims to support the full specimen lifecycle from gathering the material to accessioning in the herbarium, to publishing data in GBIF and beyond. Persistent identifiers for the Digital Specimens will improve specimen data citation and provide for connecting the specimen data with other data in the emerging web of scientific data. In my talk I will present the roadmap for the DiSSCo digital specimen infrastructure and how this will affect field work, specimen curation and research in the coming years.

# **Building pollinator resources in urban areas - pollinator friendly plants for different situations**

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Today more than half of the world's population live in cities. Urban greenspace has become an import habitat for native biodiversity. Huge potential exists in cities to provide refuges for insect, plant and animal biodiversity. Urban spaces can genuinely help with the biodiversity crisis through careful management, maintenance and selection of species. Urban green space and gardens have been found to support lots of pollinators, with studies in the UK highlighting that they can hold and support as many as those in the surrounding rural areas. This mean our gardens and natural and semi natural green spaces have an increasing important role to play in pollinator conservation.

Irelands biodiversity is categorised as "bad" with 85% of our important EU habitats classified as "bad". One particular group of Irish species is under sever threat, the bees with over one third of species threatened with extinction. Irelands capital city is Dublin habitat types have been mapped and categorised as part of the Dublin City Biodiversity Action Plan. Surprisingly Dublin is a very "green" city with less than half (43%) of the area consisting of built surfaces and a third of the city comprises of residential gardens (28%).

With all these aspects in mind this study set out to investigate which ornamental plant species in the horticultural trade might be most suited to urban gardens to support bees and other pollinators. Over 69,000 taxa are listed in cultivation on the Royal Horticultural Society (RHS) database. The review was carried out and species selected from plant databases (RHS), various "*perfect for pollinators*" lists, hobbyist websites, expert opinions and a review of Irish nursery stock trade catalogues. Species were further refined for use in twenty-four different urban planting situations. With suggested bee and pollinator friendly ornamental species for every type of urban green space from planters and flower beds to green roofs and all the way to industrial planting schemes at roundabouts. The potential benefit to urban bee and pollinator conservation is enormous. Using selected plant species enables everyone to take part and be involved with a sustainable solution to pollinator declines. We can be truly "green" in our considerations for urban planting schemes by providing food and shelter resources in our gardens for all species the important components of biodiversity which provide us with life on the planet.

#### Diversity and importance of arboreal plants

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Trees are unique creatures – they represent a group of long-lived plants belonging to extremely diverse systematic groups – from palms and woody ferns to conifers. They occur within highly variable ecological conditions – from the tropical rainforests to the coldest regions on the Earth (Antarctica excluded) The plenary lecture presents a survey on the diversity of arboreal plants (trees and shrubs) on the Earth with a particular reference to temperate zones. Some tree species, especially in the boreal zones, occupy large territories and have, therefore, substantial role in the formation of the environment. The forest ecosystems and even individual trees provide habitat for many other living organisms.

Trees possess diverse life-history characteristics – they are long-lived, have large size, and can rapidly colonize new territories. They are predominantly outcrossing, but possess high diversity of reproduction systems including wind-pollination and zoogamy. They are characterized by high levels of genetic diversity, and tend to accumulate new mutations at a slower rate. The lecture is an attempt to summarize the most significant peculiarities of the trees as an important part of the plant kingdom, even though not related in a natural systematic group. Finally, the numerous uses of the arboreal plants by humans are outlined, including the economic benefits of the different species, and the social functions of the forest ecosystems.

#### Film Plants of the Lamiaceae family as source of bioactive derivatives

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*Lamiaceae* is the largest family of the *Lamiales* order, including more than 7000 species typically cultivated as ornamental plants, as food and spices source or used in alternative medicine to treat various disease. A plethora of application are reported for the plant derivatives (essential oils/EO, hydro-alcoholic extract/HAE) in agriculture, cosmetic, food and pharmacy, due to the pleiotropic biological effects, related to antimicrobial, anti-inflammatory, antioxidant, and cytotoxic activities. Considering the antimicrobial activity (and insect-repellence), the reduced toxicity and the eco-compatibility, extracts from aromatic plants are also used in green conservation procedure of cultural and natural heritage.

Specifically, *Origanum vulgare* L. and *Thymus vulgaris* L. EOs and HAEs, have been assessed to counteract the bacterial and fungal colonies, able to induce biodeterioration processes on historic-artistic artefacts or in the inner part of cultural-historical importance trees.

Concerning Origanum vulgare and Thymus vulgaris essential oils, the exposure to their volatile compounds was successful applied against bacteria (Bacillus sp., Georgenia sp., Ornithinibacillus sp., and Streptococcus sp.) and fungal (Aspergillus sp. and Penicillium sp.) colonies, widespread on the surface of wooden sculptures or leather artworks. In order to enhance the antimicrobial activity of T. vulgaris, the EO has been applied in combination with the relate HAE or under vacuum condition. Furthermore, Origanum vulgare L. and Thymus vulgaris L. EOs have proven to be efficient biocides to counteract a complex biofilm, spreads under mosaic tiles and causing their detachment. Antimicrobial activity has been revealed also for Calamintha nepeta (L.) Savi, Crithmum maritimum L., essential oils although lower than that the previous two.

Conservation strategy for the historical ornamental trees of *Erithryna caffra* Thumb. (*Fabaceae*), species characterized by flowers of orange colour, has provided the combined use of EO and HAE from *Origanum vulgare* aromatic plant. This approach allows to counteract the *Bacillus* sp. and *Streptomyces* sp. (bacteria), and *Alternaria* sp., *Aspergillus* sp. and *Chaetomium* sp. (fungi) colonisations.

The chemical compounds of both EOs and HAEs used in these studies were identified by GS-MS analysis. Carvacrol, thymol and their biosynthetic precursors *p*-cymene and  $\gamma$ -terpinene are the main components.

In sustainable conservation strategies, basing on the several bioeffects and multiple modes of action against different groups of damaging pests, plant derivatives represent an efficient alternative to the harmful chemical synthetic pesticides.

### Medicinal Plants of the Bible - Past, Present, and Future

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Based on data on archaeobotany and ethnobotany of the Holy Land, a survey of the use of medicinal plants in the Holy Land throughout history as well as at the present time, and a revision of the medicinal plant of Assyria we suggest a new list of the Medicinal Plants of the Bible.

While Duke and Duke (1983, Medicinal Plants of the Bible) enumerated not less than 176 plant species as "Biblical Medicinal Plants" and Jacob (1993, Medicinal plants of the Bible: Another view) only 54, in our survey we suggest reducing that figure to 37. The overlap between Jacob's list and ours is 19 species in total. Our contribution is 18 "new" suggested Biblical Medicinal Plants. This discrepancy is due to three reasons: 1. Not less than 22 species in Jacob's list are not recognized today (Amar, 2012, Plants of the Bible) as valid Biblical plant names at all, or they are not related to specific species. 2. Several identifications from Campbell-Thompson (1949, A Dictionary of Assyrian Botany), the only Mesopotamian source used by Jacob, are no longer recognized by modern Assyriologists. 3. Several Mesopotamian plants were only recently identified in a medical context.

Only five species are mentioned directly as medicinal plants in the Bible: Fig (*Ficus carica*), Nard (*Nardostachys jatamansi*), Hyssop (*Majorana syriaca*), Balm of Gilead (*Commiphora gileadensis*) and Mandrake (*Mandragora officinarum* = M. *autumnalis*). Not less than 18 medicinal plants, additional to the Bible, are mentioned in old Jewish post-Biblical sources. Most of these plants (15) are known also in Egypt and Mesopotamia while 3 are only from Egypt. Seven of the BMP species are not mentioned in the Bible or in the old Jewish Post-Biblical literature but were recorded as medicinal plants from Egypt as well as from Mesopotamia, and it is quite logical to assume that they can be included as BMPs.

According to our survey, *all* 37 suggested BMPs are still in medical use today in the Middle East and are subjected, in the 21<sup>st</sup> century, to active research in attempts; to understand their chemical composition and/or Medical activity and/or Isolation of new compounds for new drug development.

### A review of the dye plants in Spain

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Since ancient times, plants have offered mankind a large number of natural dyes. Despite the great progress made by the chemical industry in dyeing compounds in the last two centuries, the disadvantages of their use at industrial level are also well known, especially their high environmental cost. This has led to a renewed interest in dye plants as a real, effective and more environmentally friendly alternative to chemical dyes.

Spain, in addition to having an extensive territory, has a high topographic, geographic, climatic and, of course, floristic diversity, which makes it one of the most diverse countries in the Mediterranean area in wild plant species. Along with a long history and great cultural diversity, the territory is rich in ethnobotanical data on the traditional use of the flora. In addition, and after several decades of research, Spanish ethnobotany is currently quite well studied. Within the framework of the project *Inventario Español de los Conocimientos Tradicionales Relativos a la Biodiversidad*, a group of 76 researchers affiliated to 17 national universities and other institutions have performed a review of the ethnobotanical literature in Spain (158 publications including 53 journal articles, 58 books and 30 Ph.D. thesis), covering the whole national territory and all the possible spectrum of traditional uses of plants.

In this communication, we present the most remarkable results regarding the traditional use and diversity of plants as dyers. For this purpose, after a necessary introduction on the botanical, cultural and ethnobotanical aspects of the territory, and a brief historical introduction to the use of plants as dye in Spain, results will be analyzed. More than 100 plant species have been used as dye plants in the Spanish tradition. We will highlight the most interesting results according to specific uses, territorial extension of the uses, analysis of botanical families, diversity of organs used, and possible regional or cultural preferences for certain species.

In addition to raising awareness of the current value of ethnobotany to present concrete solutions to current problems, we intend to show the great diversity of resources that can go from being used at a personal or family level to being used at an industrial level.

### Wild plants used as spices

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Natural plant resources provide various benefits to humans: food, shelter, fuel, timber etc. It is hard to imagine how many plant species were used in the past for food and as a spice. Today, with increasing consumption and the need for cultivated plant production, much of the knowledge about wild plants used as food and spices is threatened with extinction. And while using plant resources in traditional medicine is a fast-developing and economically important branch, their use as spices and herbs also deserves special attention and thorough investigation. Usually, spices and herbs are used to flavour the dishes and to provide diverse tastes, but many of them have medicinal effects as well. Therefore, it is not always possible to distinguish between the two groups. The terms spices and herbs are often used interchangeably, but if a more precise definition is applied, then the herbs will refer only to green, non-woody vegetative parts of the plants.

Collection, use, cultivation and trade of spices and herbs have long-term history and date back to antiquity and flourished in the Middle Ages, particularly after the Age of Exploration. Many species of commercial importance are cultivated, but the wild-growing plants that can be used as spices and herbs are still much more, especially in some remote and less-known places on the Earth. The plenary lecture presents a review on the use of wild plant species as spices and herbs. It is related to many factors: the species occurring in a given area, the local traditions and habits in cooking and food preparation and many others, which are discussed in detail. Practically all plant parts are being used as spices – roots and rhizomes, stems of herbaceous plants and leaves of all plants, the bark of arboreal plants, flowers and flower parts, fruits and seeds. Information is provided about the plant groups and plant families used most frequently as spices and herbs in Europe and in some other parts of the World.

### **Edible Treasures: Discovering the Riches of Wild Food Plants**

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Traditional knowledge about wild food plants is being lost at an alarming rate around the world, despite the growing interest in them among scientists and the general public. Throughout human history, wild food plants (WFPs) have been a part of diets and traditional food systems, supplying vital nutrients and bioactive substances. Traditional diets, both ancestral and modern, are recognized to provide significant health benefits. On the other hand, rural societies all over the world are being affected by globalization, which is making them more dependent on outside resources and cutting them off from their own, like wild plants or landraces. Wild food plants include a wide range of species that grow naturally in places like woods, fields, meadows, and even cities. These plants provide a substantial nutritional benefit since they typically have higher vitamin, mineral, and antioxidant content than conventionally grown foods. However, wild food plants offer unique and distinct flavors and textures that can bring a sense of novelty and adventure to our meals. The integration of wild food plants into our diets promotes a seasonal and locally-sourced method of consumption, which cultivates a stronger bond with the environment. Many wild food plants have medicinal properties and have been used in traditional herbal medicine for centuries; these plants offer potential health benefits beyond mere nutrition. Wild food plants help us appreciate the wealth and diversity of nature, encouraging conservation and sustainable living.

#### Saffron (Crocus sativus, Iridaceae) in Ocular Diseases

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*Crocus sativus* L. from *Iridaceae* family is a pricy plant in the world's trade. The stigmas of the saffron flowers are known as a dye and spice since ancient times. Traditionally, dried stigmas, saffron, are also used as a medicine in the treatment of different diseases such as antihypertensive, antitussive, anticonvulsant, antigenototoxic and antioxidant, cytotoxic effects, anxiolytic aphrodisiac, antinociceptive, antidepressant, antiinflammatory, and relaxant activity.

Macular degeneration is the main cause for elderly's oculars and carotenoids delay and help to treatment of mild and moderate macular degeneration. Saffron contains different class of compounds which are mainly terpenoids, carotenoids, and phenolics. Crocins, crocetins, and picrocrosin were isolated from saffron. In last decade, many produced pharmaceutical products for retinal deformations and age-related macular degeneration contain carotenoids as active constitutions. This study summarizes the carotenoid derivatives of saffron and discusses their beneficial effects on the oculars diseases.

## Pharmacognostic characterization of *Diplotaxis tenuifolia (Brassicaceae)* from Campania and Sicily (Southern Italy)

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*Diplotaxis tenuifolia* (L.) DC., known as perennial wall-rocket, and called "rucola selvatica" in Italy, is a herbaceous plant traditionally used in the Mediterranean area mainly as a wild salad, but also appreciated for its beneficial properties.

Leaves have in fact notable nutritional properties related to the richness in glucosinolates and antioxidant compounds, such as vitamin C and phenolic compounds. Some uses have been reported in the traditional medicine of Southern Italy, such as a laxative, for the treatment of liver problems and as an aphrodisiac, but also as a diuretic, and against myalgia and dental problems. In addition, a cosmetic use to promote hair growth has been referred for Sardinia, and the species has also been cited for its mucolytic properties.

However, to date there is a paucity of studies on the potential medicinal properties of this species, which could pave the way for new nutraceutical and phytotherapeutic applications. Therefore, the aim of our study was to analyse the anatomical and micromorphological features of the leaves of the plant and evaluate the phytochemical profile and the antioxidant activity of a hydroalcoholic extract obtained from leaves of two populations, growing in Campania (DtC) and in Sicily (DtS), respectively.

# Anthriscus sylvestris (Apiaceae): Phytochemical and pharmacological approach

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Anthriscus sylvestris (L.) Hoffm, also known as cow parsley or wild chervil, is a wildgrowing biennial or perennial plant from Apiaceae family, belonging to the same tribe (Scandiceae) as better-known edible and medicinal plants such as garden angelica, cumin and carrot. A. sylvestris has no use in Western countries and USA, where is considered a nuisance weed due to its high invasiveness. However, in China, Japan and Korea it is a part of both diet and traditional medicine as antipyretic and analgesic, cough remedy, diuretic, hematinic, tonic, digestive, antihypertensive etc. These therapeutic effects are the results of the presence numerous secondary bioactive molecules such as lignans, phenylpropanoids, coumarins, flavonoids, polyacetylenes and terpenoids. Therefore, it is reasonable to assume that A. sylvestris is an attractive target for phytochemical and biochemical research. Here we provide an overview of chemical and biological studies of wild-growing A. sylvestris from Serbia. Phytochemical study revealed that phenolics are the most abundant compounds, with lignans as dominant and the most important secondary biomolecules of A. sylvestris, due to their biological activities, most notable potent cytotoxicity. Our systematic screening, based on HPLC-MS/MS-guided chromatographic enrichment and purification, resulted in full or partial identification of 46 lignans, including 19 previously unknown. The most abundant classes were found to be aryltetralins and saturated and unsaturated dibenzylbutyrolactones, typically bearing guaiacyl (3-methoxy-4-hydroxy), veratryl (3,4-dimethoxy), piperonyl (3,4-methylenedioxy) or syringyl (3,4,5-trimethoxy) substitution. By depleting the more abundant lignans, we managed to isolate or at least make detectable nearly 50 minor compounds, mostly dibenzylbutyrolactones and aryltetralins, but also dibenzylbutandiols, tetrahydrofurans and arylnaphthalenes, all bearing different combinations of several common substitution patterns. Beside lignans, the most abundant phenolics were phenylpropanoids, especially mono- and dicaffeoylquinic acids. Phenylpropanoids based on O-prenylated coniferol have never before been reported in Anthriscus spp. Employing LC with MS/MS, derivatives of several flavonoid aglycones - luteolin, chrysoeriol, apigenin and quercetin, were identified. HC-GC-MS was used to obtain volatile profile of individual parts of the plant. The dominant components were monoterpenes (with ~15 %  $\alpha$ -pinene) but also alkanes (~20 % *n*-nonane), Different fractions of A. sylvestris were subjected to several in vitro and ex vivo assays for cytotoxic, anti-inflammatory and antioxidant activity. The structure-activity relationship (SAR) and possible mechanism of action is discussed. The results of this study confirm our hypothesis that A. sylvestris deserves attention as the source of potent phytopharmaceuticals.

## Phytochemical constituents, biological activities, and health-promoting effects of Sicilian wild artichokes

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The Sicilian wild artichokes, *Cynara cardunculus* L. subsp. *cardunculus*, *C. cardunculus* subsp. *flavescens* Wiklund, *C. cardunculus* subsp. *zingaroensis* (Raimondo & Domina) Raimondo & Domina, are thorny species related with the cultivated artichokes [*C. scolymus* L., *C. cardunculus* var. *scolymus* (L.) Fiori, *C. cardunculus* subsp. *scolymus* (L.) Hegi] present in Sicily with different ecotypes.

*Cynara cardunculus* subsp. *cardunculus*, synonym *C. cardunculus* L. var. *sylvestris* (Lam.) Fiori, is the main taxon who has been appreciated for its culinary qualities for centuries. In nature it grows in various types of environments and soils, including rocky hillsides, dry meadows, showing great adaptability to different climatic conditions, and has a long history of medicinal uses due to the presence of phenolic compounds and flavonoids. Aerial parts have been utilized for their therapeutic properties on the digestive system and liver. The leaf aqueous extract of wild artichokes grown in Siracusa (Italy) was investigated in this research project for its phenolic profile and cell-free antioxidant activities. Following this, the crude extract was assessed for its protective properties and potential oxidative stress inhibitory effects in HepG2 cells treated with free fatty acids to simulate the early stage of mild steatosis in humans, as well as for its possible chemotherapeutic activities in Caco-2 cells as an *in vitro* model of colorectal carcinoma.

Several phenolic acids (caffeoylquinic acids) and flavonoids (derivatives of luteolin and apigenin) were found by HPLC-DAD analysis and a promising antioxidant power was also demonstrated by the DPPH assay ( $IC_{50} = 20.04 \ 2.52 \ \mu g/mL$ ). The crude extract was found to be capable of reversing the damage caused by FFA exposure in HepG2 cells by restoring cell viability, reducing oxidative stress by suppressing ROS and LOOH, and raising RSH levels through the modulation of mRNA expression levels of antioxidant defense markers and of inflammatory cytokines. On Caco-2 cells, wild artichokes extract reduced cell viability in a dose-dependent manner by interfering with protein expression levels of antioxidant defense markers (HO-1) and pro-apoptotic factors (Bax and p53).

These findings add new evidence to the health benefits of this taxon of the *Cynara* genus, suggesting that the total phytocomplex present in the leaf is a valuable source of bioactive compounds that not only effectively counter hepatic oxidative stress but also exerts chemotherapeutic effects on colon cancer cells. This underutilized species has the potential to be a sustainable economic resource for the natural health product sector because it can efficiently grow on stony, poor soils without the addition of water.

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# Utilization of endophytic fungi for biotransformation to obtain bioactive compounds

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Biotransformation is defined as biochemical reactions of living systems or their components (enzymes) to modify molecules. It has been suggested as an effective tool in obtaining compounds that are difficult to prepare by synthetic methods. One of the research topics of our team is the modification of plant secondary compounds by the same plant's endophytic fungi and the investigation of metabolites' bioactivity toward cellular aging and age-related diseases.

The overriding basis of our interest in aging is that age-related diseases have become an economic and psychological burden for societies. Some significant hallmarks of aging are loss of proteostasis, telomere attrition, and increased ROS. These alterations lead to age-related disorders such as cardiovascular diseases, neurodegeneration, and cancer. Thus, there is a substantial demand for new prevention and treatment alternatives. With their unique chemical diversity, natural products have been proposed as potential agents (i.e., resveratrol, cycloastragenol, curcumin, quercetin) for cellular regeneration, healthy aging, and age-related diseases.

In the first part of my presentation, I will focus on a well-known anti-aging agent (cycloastragenol) and its metabolites deriving from our biotransformation studies using endophytic fungi. The effects of the metabolites in cellular senescence in terms of NRF-2/proteasome/telomerase systems will be described. Secondly, I will share several new metabolites possessing cytoprotective activities, especially neuroprotective ones.

We thank the Pharmaceutical Sciences Research Centre (FABAL, Ege University, Faculty of Pharmacy) for equipment support. We are very grateful to Bionorm Natural Products for providing cyclocephagenol and cycloastragenol. This study was supported by TÜBİTAK (Project number: 119Z086) and Ege University Research Fund (Grant number: GAP-23729).

# Enhancement of ancient fruit varieties registered in the regional repertoires through the study of their health properties

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The valorisation of local agri-food products by characterizing their healthy properties is essential in this historical moment. In fact, typical local food products play a significant economic role, particularly in the Mediterranean area, where food processes and products are very diversified. This diversification determines the agricultural biodiversity. The enhancement of these processes and products, often obtained from quality brands, ie DOP, IGP, STG, TAP, can be further pursued through their nutritional and nutraceutical characterization. Knowledge of the healthy characteristics of particular local foods can be used for their diversification and marketing. The regions of the Mediterranean area including Emilia Romagna are rich in local and traditional agri-food products. Since the 1950s, also in Italy as in the rest of the Mediterranean area, intensive fruit growing has developed, based on specialized orchards and the ancient varieties have gradually become a memory: of the varietal heritage existing between the 17th and 18th centuries, painted by the Medici painter Bartolomeo Bimbi, only 17 types of pear trees out of the 115 existing ones have come down to us. In this context, we found it interesting to study the ancient variety of pear named "Cocomerina" in order to characterise it from the point of view of health promotion, with the aim of pursuing its diversification and valorisation. In our studies, we focused on two ecotypes of "Cocomerina" pear whose main feature is the reddish or red color pulp. In addition, the comparison with the nutritional and nutraceutical properties of different ancient pear varieties was also studied.

Our results confirm the importance of further enhancing the fruit trees which seem to have been forgotten, taking into account that the old varieties are often the most suitable for agriculture with low environmental impact. It is evident that with the climate changes, plants characterized by high adaptability and resistance are needed to counteract the biotic (caused by fungi, bacteria, nematodes and various insects) and abiotic stress (dependent on water availability and water, light and temperature quality). These characteristics have been demonstrated for all the "forgotten fruits" already studied. Based on the above considerations, in order to meet the growing demand for healthy food, free from damage and harmful organisms, it is desirable to recover, improve and promote the cultivation of these fruits.

In the future, we will have to face a new study in order to definitively identify the compounds biologically active in "Cocomerina" pear. In addition, the bioactivity of this pear juice and different extracts, will should be tested with in vivo model.

## Antioxidant and anti-inflammatory activity of ancient apple varieties from central Italy

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The Mediterranean region is considered one of the world's richest areas in terms of biodiversity.

As a result of a complex evolutionary process that has evolved in response to a territory's characteristics and relationships between species, biodiversity is the primary source of the link between environmental and cultural diversity.

Preserving biodiversity means saving a genetic heritage of exceptional environmental, economic, social and cultural value, the latter being rooted in ancient rural and artisanal knowledge.

In recent years, due to their health, environmental and economic benefits, so-called 'ancient fruits' have attracted particular interest.

The recovery and subsequent study of some ancient cultivars have helped identify them as potential sources of secondary metabolites, and many of these fruits belong to the genus *Malus (Rosaceae)*.

This genus comprises some 35 species, of which *Malus domestica* (Suckow) Borkh is undoubtedly the best known, as its fruits has been part of human history since ancient times.

*M. domestica,* which produces the apple of our tables, derives from ancient and continuous hybridisation between different species of the genus *Malus*. Its progenitors, as *M. sieversii* (Ledeb.) M. Roem., *M. dasyphylla* Borkh., *M. pumila* Mill., *M. sylvestris* (L.) Mill., *M. orientalis* Uglitzk., *M. baccata* (L.) Borkh., *M. mandshurica* (Maxim.) Kom. ex Skvortsov, are generally known as 'wild apples'. The main ancestral species has been identified in *M. sieversii*, which grows in the last primary forests of central Asia and is experiencing a severe decline due to habitat loss.

Apples have a high nutritional value, they are an important source of polyphenols, anthocyanins, vitamins, minerals, and based on their properties, they are rightly included in the category of "functional foods".

In this work, we summarised and compared the results we have obtained from the study of ancient apple varieties and cultivars, which are an integral part of the food and agricultural culture of the areas in which they originate.

We have evaluated the total polyphenol and anthocyanin content, and also the antioxidant and anti-inflammatory activities of crude juices and extracts from three different apple varieties: "Abbondanza" (clone with red pulp from Romagna region), "Pelingo" and "Rosa in pietra" (from Marche region). The first two are apples characterised by intense red skin and flesh.

The results obtained make us increasingly convinced that intensifying the cultivation of these apple varieties, without altering their characteristics, could also be beneficial from a purely health perspective.

# Recent introduction into cultivation of wild plants in the Iberian Peninsula

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Amongst the 6.176 Angiosperms native to the Iberian Peninsula and the Balearic Islands, over 200 are progenitors of widely cultivated plants. Some were introduced into cultivation in the Near East already in the Neolithic; this is the case, by instance, of *Linum bienne* Mill. (the progenitor of the flax, *Linum usitatissimum* L.) and *Lathyrus sativus* L. (grass pea). Some other were introduced into cultivation latter, in the Bronze Age, also in the Near East, including the three more characteristic Mediterranean woody crops: *Ficus carica* L. (fig), *Vitis vinifera* L. (grape vine) and *Olea europaea* L. (olive), and some herbaceous crops, as, for instance, *Trigonella foenum-graecum* L. (fenugreek). Several are cultivated since Greek and Roman times, as it is the case of *Corylus avellana* L. (hazel), *Malus sylvestris* (L.) Mill., *Laurus nobilis* L. (bay laurel), *Brassica oleracea* L. (cabbage), *Apium graveolens* L. (celery) and many other. And a few have been introduced into cultivation in recent times, such as *Beta maritima* L. (the progenitor of *B. vulgaris* L., beet, beetroot, sugarbeet) and *Lupinus luteus* L. (yellow lupin).

But four species which, together with several hundred wild species have been gathered for food or as pot-plants since ancient times, have been recently introduced into cultivation in Spain and are now harvested and commercialized as new crops. Of them, the cultivation of *Borago officinalis* L. (borage), a most popular dish of the Aragonese cuisine, started at the beginning of the XIX century; *Scolymus hispanicus* L. (golden thistle), has been introduced into cultivation in Cádiz province (S Spain) at about 20 years ago, and *Silene vulgaris* (Moench) Garcke (bladder campion) is currently in the phase of agronomic studies for its introduction into cultivation. *Prunus spinosa* L. (blackthorn), a shrub up to 5 (-6) m tall, which fruits are the base of an important liquor industry (which product is commercialized with the name of "pacharán"), is cultivated in the province of Navarra (N Spain) since 1987.

### Conservation and use of wild relatives of cereals and food legumes

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Cereals and food legumes remain the major components of human diets worldwide. In the Central, West Asia and North Africa, wheat, barley, chickpea, lentil and faba bean are providing more than 65% of energy and proteins in human consumption. Improved varieties are major inputs to increase the productivity and quality of these crops and to overcome the challenges imposed by climate change. Moreover, new varieties with increased nutritional quality are one of the most important tools to achieve nutritional security in the Global South. Genetic resources and mainly landraces and wild relative species are key for ensuring continuous genetic gains and introgress genes of tolerance and resistance to major abiotic and biotic stresses.

ICARDA has a global mandate for the genetic improvement of barley, lentil, faba bean and grass pea and shared mandates for the improvement of chickpea and wheat with other international centers. The genebank of ICARDA has important collections totalling more than 150,000 accessions of cereals, food legumes and temperate forages including unique and important holdings of wild species, most of which are collected from three Vavilovian centers of diversity. laying in CWANA region. This region is renowned for its remarkable biological diversity and stands out as a global center for plant genetic resources accounting for 41% of the world's essential crop wild relative (CWR) taxa. Crop wild relatives are increasingly used in enriching the genetic diversity of cereals and food legumes through strengthening of pre-breeding activities. In fact, the evaluation of sets of accessions of wild species from genepools 1 and 2 has allowed the identification of sources of high contents of micro-elements for crop biofortification.

Successful interspecific crosses were done in case of barley with *Hordeum bulbosum*, in case of wheat with *Triticum urartu* and *T. boeticum* and several Aegilops species, in case of grass pea with five *Lathyrus* species and in case of chickpea and lentil with few *Cicer reticulatum*, *C. echinospermum* and *C. judaicum* and *Lens orientalis* and *L. ervoides*. The derived lines from interspecific crosses showed improvements in yield under both drought and optimum conditions, resistance to rusts, septoria, tan spot and Hessian fly in both bread and durum wheats; resistance to powdery mildew, scald, net blotch and rusts and high  $\beta$ -glucans and yield stability in barley; higher yields, early maturity,tolerance to ascochyta blight, fusarium and orbanche, higher contents of iron, zinc and selenium and tolerance to herbicides in case of lentil and chickpea and low ODAP content and orobanche resistance in grass pea. The derived germplasm have also shown enchanced tolerance to drought and heat.

The international nurseries distributed by ICARDA include the lines derived from interspeciefic crosses representing 20-30% in case of durum wheat, 10% in case of barley and 10-30% in case of chcickpea and lentille. Some of these lines have already been released by national programs in Morocco, Lebanon, India and beyond. Recently, two lentil varieties (Jammu Lentil 144 and Jammu Lentil 71) from the use of CWR (*Lens orientalis*) have been released in India.

# Introduction into cultivation of the Balkan endemic *Sideritis scardica* (*Lamiaceae*)

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*Sideritis scardica* Griseb. (*Lamiaceae*) is a short living perennial herb. This Balkan endemic is a valuable medicinal plant, which grows in the higher parts of the mountains of Bulgaria, Albania, Greece, and Macedonia. The distribution is rather fragmented. *S. scardica* populations in Greece are the most preserved natural ones within the range of the species. According to Global and European regional assessment *S. scardica* is considered Near Threatened (NT) but in Bulgaria its IUCN category is Endangered (EN). In the last decades this species became particularly popular medicinal and table tea plant in Bulgaria and its populations suffer from significant anthropopressure. The solution of this problem is cultivation and *S. scardica* is successfully grown comersially in many places in Bulgaria lately.

*Sideritis scardica* is propagated by seed. Under cultivation vegetative reproduction is possible by rhizome division, but there has been no evidence of spontaneous vegetative reproduction. Therefore seed is needed not only to preserve the wild populations, but to produce seedling material for *in vitro* (biothechnologically) and field cultivation. Consequently it is crucial to know the factors that control the seed production and the pollination in particular, regarding the global pollinators' decline. The field experiments with enclosed flowers reveal that spontaneous self-pollination does not occur and insect pollen vectors are required. Our investigations in six study sites (three *in situ* and two *ex situ*) in the Rhodopes and one *ex situ* study site in the foothills of Pirin Mts reveal that the pollinators of *S. scardica* are polylectic insects such as bumblebees and carpenter bees (both *in situ* and *ex situ*) as well as honeybees (only in the big cultivated fields).

The protection of the wild populations of *S. scardica* as well as its successful cultivation depends on the protection of its pollinators.

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## Applying genomic tools in the study of Crop Wild Relatives in the south Levant

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The south Levant, as part of the Fertile Crescent, is considered as a hot spot of several important crop wild relatives (CWR). Environmental gradients in this region offer opportunity for eco-evolutionary processes that lead to genetic sub-clustering between populations/ecotypes. Thus, CWR and crop landraces might possess valuable adaptive diversity, which can contribute to safeguard food resources. Genome scan methodologies offer opportunity to characterize this unique germplasm of the south Levant. Applying genomic sequencing technologies (whole genome sequencing and GBS) assisted us to characterize adaptive genetic differentiation in populations of wild arugula (*Eruca sativa*) and local wild radishes (*Raphanus raphanistrum* and *R. pugioniformis*), as well as to identify loci under selection. In *Olea europaea*, microsatellites markers were used to identify the 'founder genetic stock' of *O. europaea* subsp. *europaea* var. *sylvestris*, cultivar diversity, and to infer about the influence of agricultural activity on genetic structure of populations of wild olive trees. Considering the rapid decline in natural habitats in this region, the results of these studies provide tools for planning and prioritization the conservation efforts in Israel.

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# Taxonomic and genetic diversity of the Sicilian relatives of azarole (*Crataegus azarolus, Rosaceae*)

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The azarole (Crataegus azarolus L.) in Sicily has been cultivated since ancient time in the two chromatic forms of the fruits, respectively yellowish [C. azarolus var. chlorocarpa (Moris) K.I. Chr.] and reddish [C. azarolus var. pontica (K. Koch) K.I. Chr.]. Both forms are also found spontaneously. Alongside of the spontaneous populations of the cultivated species, various forms recur, partly referable to well-defined taxa, partly to hybrids often of unclear origin. Not all wild forms can be considered related to C. azarolus and therefore can be considered relatives of this species which, according to the authors, may have a wild ancestor in C. aronia (L.) Bosc ex DC. ( $\equiv C.$  azarolus var. aronia L.), very widespread in Sicily in a small area of the ancient territory of Erice, or in the coastal plain of S. Vito Lo Capo in the province of Trapani (N-W Sicily). These populations are morphologically distinct and live together in the same area. They may relate to both C. azarolus var. azarolus and with C. aronia. To the congener taxa recognized in the literature we two new specific entities typical of the Erice area should be added. Another case subject to various taxonomic interpretations is represented by Crataegus laciniata Ucria synonymized by some with C. orientalis (Mill.) M. Bieb. and then re-evaluated in the original specific taxon established by Bernardino ab Ucria at the end of the 18th century. In consideration of the recurring taxonomic diversity in Sicily - distinguished exclusively at the morphological level - a genetic investigation was carried out including all the recognized and published taxa. SSR (Simple Sequence Repeat) molecular markers isolated in species belonging to the Rosaceae family (apple, pear), reported to be transferable between related species, were used in this preliminary study, to better characterize them and to support the taxonomic interpretations. The isolation of Crataegus-specific SSRs would be desirable to better complement and support the morphological characterization.

# Taxonomy, ecology and distribution of the Sicilian relatives of artichoke (*Cynara scolymus, Asteraceae*)

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*Cynara* L. (*Asteraceae*, *Carduoideae*) is a genus native of the Mediterranean and the Irano-Turanian regions. It includes about 10 species and among these only *Cynara scolymus* L. is widely cultivated; this taxon is often treated as a subspecies [*C. cardunculus* subsp. *scolymus* (L.) Hayek] or as variety [*C. cardunculus* var. *scolymus* (L.) Fiori] or most recently as synonymous (see <u>https://wfoplantlist.org</u>). Regardless to the systematic classification, some other species and subspecies are counted among the wild relatives of the cultivated taxon. Among the wild relatives of *C. scolymus* a prominent place has always been occupied by *C. cardunculus* L., a wild species widely accepted as the ancestor of the cultivated forms.

The artichoke (C. scolvmus) is one of the oldest vegetables; in Italy it was mostly selected and cultivated especially in Roman times and has characterized cooking recipes in the last two thousand years and beyond. In Sicily, the artichoke always had an economic interest; it includes several cultivars each typical of a countryside area where his potential is highest like the 'Spinoso Palermitano' or 'Spinoso di Cerda', the 'Vagghiàrdu' or 'Violetto di Niscemi', the 'Violetto Catanese' (or 'Ramacchese'), the 'Violetto Siracusano' (or ' Violetto di Lentini'), the 'Spinoso di Menfi'. In addition, different ecotypes are widespread cultivated in Sicily, often propagated by vegetative means, a germplasm as well useful and worthy of being recorded and preserved. In Palermo, where the artichoke most distinguishes street food, alongside the thorny violet – the most renowned and commercially expensive - three other forms are cultivated, partly unarmed, two of which have flower heads very similar to the nominal subspecies of C. cardunculus and of which the small flower heads are used, boiled in lightly salted wate. The wild thistle (C. cardunculus) is historically harvested and eaten during the winter until spring as boiled and/or fried and (or in batter) vegetable and often characterized the historical dishes on the tables of San Giuseppe. Moreover, in some disadvantaged areas (e.g. Cammarata, Valle dell'Inferno) there was a tradition in late spring to eat the heads of the wild thistle after patiently removing the thorns.

The wild ancestor *C. cardunculus* subsp. *cardunculus* in Sicily is widely distributed mainly in the grazed clay soils, from the coastal area to the mid-mountain. The species includes other two subspecies: *C. cardunculus* subsp. *zingaroenesis* (Raimondo & Domina) Raimondo & Domina growing in the western part of the island; *C. cardunculus* subsp. *flavescens* Wiklund, native of the Iberian Peninsula, Canary Islands and North Africa, has been reported in Sicily for an inland hill of Caltanissetta province, but according to our investigations it occurs in a wider area.

Within the genus *Cynara*, in the Mediterranean region, natural hybrids are also known and among these one recently described in Spain (*C.* ×*gaditana* Blanca & Sánch. Carr.; *C.* ×*pacensis* F.M. Vázquez). Also in Sicily, a limited wild population found in the Monti Sicani area has a hybrid character, with evident intermediate characters between some cultivated forms of *C. scolymus* and the widespread *C. cardunculus* subsp. *cardunculus*.

In conclusion, in Sicily, the artichoke (*C. scolymus*) would have as wild ancestor *C. cardunculus* subsp. *cardunculus* and as wild relatives occur: *C. cardunculus* subsp. *zingaroensis* and *C. cardunculus* subsp. *flavescens*.

# Cataloguing the alien flora of north-western Mediterranean basin: recent efforts and future prospects

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The Mediterranean basin, in particular its north-western part, has become (and will continue to be) one of the areas of the planet most affected by biological invasions, due to a series of reasons that include: (1) a very profound modification of their natural habitats, (2) a great topographic and climatic diversity, (3) it is one of the world's main commercial and transportation hubs, (4) it is one of the world's main touristic hotspots, and (5) it will be greatly affected by climatic change. The biological invasions are even more worrisome when one takes into account that north-western Mediterranean Basin is very rich in plant diversity, with plenty of narrow endemics (that are per se very vulnerable to any human impact) and populations of high genetic singularity.

Among the most important tools for the prevention, control and management of biological invasions, the cataloguing of alien species plays a central role. However, and despite the native plant richness, the alien flora of this region has not been studied enough compared to other areas such as central and northern Europe. This communication will review some of the main efforts to catalog the alien flora of the north-west of the Mediterranean basin during the last decade. Along the communication we will present our own work as well other research carried on in this part of the basin, and will include various geographical scales, from studies of certain areas of a few km<sup>2</sup> to regions of thousands of km<sup>2</sup>, great diversity of environments (e.g. purely urban areas, mountainous areas, coastal regions) and studies focused on certain groups of highly invasive plants (e.g. succulents).

## Invasive alien plants of European Union concern in Italy: distribution and threats

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Invasive alien species are among the main global drivers of biodiversity loss, posing major challenges to nature conservation in Italy and in its network of protected areas. Furthermore, they causes significant economic loss to agriculture and forestry. In the European Union a dedicated regulation "on the prevention and management of the introduction and spread of invasive alien species" - Regulation (EU) no. 1143/2014 - was adopted and came in force in 2015. The Regulation identifies a list of invasive alien species of "Union concern" whose introduction or spread threaten or negatively impact biodiversity and ecosystem services and such adverse impacts require concerted actions at the European Union level. The identification of these invasive alien plants follows a strict administrative and technical procedure that, besides documenting with a standard and peer reviewed risk assessment document the species pressure on natural ecosystems, verifies their capability of establishing viable populations and spreading under current environmental conditions and in foreseeable climate change scenarios.

Among the 41 invasive alien plant species of Union concern that are actually regulated, around 50% of them are already present in Italy with different degrees of naturalization and invasiveness. Importantly, some of them are quite widespread, requiring long-term persistent controls that are likely to only mitigate further spread of prevent re-invasion in areas were local eradications took place. However, given the limited available resource for invasive alien species management all efforts should be preferentially addressed to tackle invasive alien plants in the Italian Mediterranean and Continental bioregions and on small Italian islands. Furthermore, considering that climate change scenarios predict an increment of two factors that at present limit invasive alien plants altitudinal spread (e.g., temperatures and precipitations), an expansion in the Alpine region could be also expected and should be prevented. Local authorities and the managers of protected areas should pay particular attention to the presence of communication infrastructures as well as to the local landscape and land use changes which may have crucial roles in promoting or preventing plant invasions. The staff of protected areas should conduct periodical field surveys in particular on areas with high/medium invasion risk to record the presence of alien species and their invasion stage.

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## Changing perceptions of invasive plants in Serbia

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The first data on the occurrence and presence of alien species on the territory of present-day Serbia date from the end of the 18th century, followed by the recording of changes in their distribution at the beginning of the 19th century. They refer exclusively to the northern part of Serbia (Vojvodina Province), which is part of the Pannonian Plain, where there are no natural barriers to the invasion of these species from Central Europe. In the older literature, alien species were assumed to occur mainly in the Pannonian Plain in Serbia and to be widespread there. Later data show that they have also invaded the hilly and mountainous part of the Balkans in Serbia and even protected areas. Until twenty years ago, no special attention was paid to the systematic study of invasive alien species and data on them are mostly sparse, while the distribution and dispersal pathways have been followed in detail only for a few species. The previous knowledge about this group of plants in Serbia has not yet been revised and adequately assessed. However, awareness of the extraordinary importance of knowledge about alien, especially invasive species for the protection and conservation of the diversity of native flora and vegetation led to a change in perception and thus to a new approach to research and interpretation of the significance of the results. The re-evaluation of published data as well as the information from newly discovered herbariums from the 18th and 19th centuries, form the basis for the review of the number of alien species, the time of introduction, the directions and dispersal modes, the spatial distribution and the invasiveness of these plants, which will have scientific and practical significance. It is already clear that the assumption that the number of invasive plants is significantly higher in the Pannonian Plain than in the mountainous Balkan part of Serbia is not justified. The distribution shows that the number of invasive species is relatively balanced in the respective habitats, e.g. grass habitats in the lowlands versus meadows and pastures in the mountains, while riparian and marsh habitats are most threatened in Serbia. The report provides an analytical overview of the current state of knowledge on invasive alien plant species in Serbia.

# An overview on alien macrophytes in Sicilian Marine Protected Areas (southern Mediterranean Sea)

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Non-indigenous species (NIS) are widely recognized as one of the main drivers of global change, threatening native biodiversity, ecosystem functioning and services. NIS may in time become invasive (Invasive Alien Species – IAS), determining significant environmental impacts, such as biodiversity loss and ecosystem services degradation. Sicilian Islands and all the Marine Protected Areas (MPAs) they host, are notably vulnerable to biological invasions, due to their strategic position at the crossroads between the south-western and eastern Mediterranean Sea, by virtue of the intense maritime traffic. The impact of NIS on marine habitats within MPAs, whose major aim is biodiversity conservation, can be significant, even highly detrimental. Therefore, monitoring NIS distribution is crucial in these areas for planning effective conservation strategies.

To date, a total of 25 macrophytes was recorded for the Sicilian MPAs, belonging to the following taxonomic groups: Rhodophyta (15), Ochrophyta (6), Chlorophyta (3) and Tracheophyta (1). Pelagian Islands, Plemmirio and Ciclopi MPAs, located in the Strait of Sicily and the Ionian Sea respectively, registered the highest number (13), followed by the Egadi Islands MPA (12), located in the Tyrrhenian Sea. The red alga Asparagopsis armata Harvey and the green alga Caulerpa cylindracea Sonder were the most frequently recorded species. The presence of C. cylindracea in all the Sicilian MPAs, confirms the invasiveness of this species, which is able to compete with native macrophytes, and may also change native benthic communities. Despite their fundamental role in the conservation of marine biodiversity, MPAs are not immune to NIS, evidencing their vulnerability to this phenomenon and confirming that protection does not hinder the introduction and spreading of NIS. Maritime traffic is certainly the main vector for the introduction and spread of NIS in the Mediterranean Sea. Since MPAs are popular touristic destinations, the expansion of NIS by recreational vessels represents a risk for MPAs. NIS will continue to increase in the Mediterranean Sea, so implementing effective policies and management action is urgently required within MPAs but also in their surrounding areas in order to manage continuous spillover effects. The creation of permanent observatories and alarm systems, able to early detect new introductions and to follow the spread of species already present might be an effective tool in the management of present and future introductions of NIS in MPAs. To this end, the citizen science initiatives may provide a valuable contribution in raising awareness, collect data, and also flagging early-warning signs.

## LIFE medCLIFFS: towards an integrative management of invasive plant species in the Costa Brava (north-east Iberian Peninsula)

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LIFE medCLIFFS (https://lifemedcliffs.org/en/) is a project financed by the LIFE Programme of the European Union, with a duration of five years and which focuses on the Costa Brava and the Natural Park of Cap de Creus, one of the most affected areas with coastal cliffs in Catalonia (north-east Iberian Peninsula) by biological invasions. Project activities range from preventive to eradication actions and, with the same level of importance, social awareness and dissemination activities. In this sense, it involves public administrations, scientists, volunteers and the ornamental plant production sector among its partners, and seeks the engagement of individuals and local entities.

The actions are basically focused on the early detection and eradication of five invasive plant taxa: Opuntia ficus-indica and O. stricta, Carpobrotus acinaciformis, C. edulis (and their possible hybrids) and Gazania rigens, which should also serve to reduce their impact on sensitive areas that harbour threatened (and narrow endemic) plant species such as Limonium geronense, L. tremolsii and Seseli farrenyi. In parallel, participatory networks have been created for the early detection and monitoring of invasive or potentially invasive alien flora, made up of observers and volunteers who provide key information to update and calibrate automatic an invasion risk assessment system (RISKMAPR; https://besjournals.onlinelibrary.wiley.com/doi/full/10.1111/2041-210X.13284).

In addition, since horticulture and the use of ornamental plants are the main cause of the establishment of invasive plants, the following specific actions are also planned: drafting of a code of conduct including a list of invasive or potentially invasive species to avoid, and a list of native or non-invasive alternatives, as well as the creating a quality label for suppliers and large users such as town halls and other public administrations.

## Invasive alien plants in Menorca (Balearic Islands, Spain): characterization, management experiences and proposals for a sustainable control

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The proliferation of invasive exotic species is considered one of the most important environmental threats. The impact of its expansion is not restricted only to the natural environment, its negative consequences also affect other areas such as the economy or human health.

Menorca, like other island territories, is particularly sensitive to the negative effects of invasive plants. A well-known experience on the island is the proliferation of *Carpobrotus*, which had serious effects on the coastal endemic flora. For this reason, the LIFE FLORA Menorca project (2001–2004) had as one of its objectives the eradication of this invasive plant.

Prevention is the best tool to avoid and control the negative effects of invasive plants. If you are aware of their existence, their distribution and, even more, if you can foresee their introduction or proliferation, then their management is much easier. In all of this the aspects such as knowledge of the territory, the trends in anthropogenic uses of plants, the plant species that circulate in commercial areas, the behaviour of non-native plants in other similar territories, etc., play an important role. For example, if the main reason for introducing exotic plants is their use as ornamentals, knowing the trends in gardening, which species or groups of species are being promoted, helps to know which invasive or potentially invasive plants are being promoted.

The first basic information for the management of invasive plants is to have a catalogue or list that has associated data on their origin, cause of introduction, mechanisms of dispersion, behavior, etc. Based on field observations, 74 non-native plant taxa have been identified for their invasive behavior, some still in an incipient phase of the expansion process.

The analysis of the geographical origin, life form, cause of introduction, method of reproduction and vectors of dispersion within the territory, provides useful information on its management. In this group of 74 taxa, those of American origin predominate (57%) and the main reason for introduction is for their use as ornamentals (65%). Up to 16 taxa have changed the cause of dispersal once introduced. For example, 7 taxa that were introduced as ornamentals, now behave as contaminants. Regarding the method of reproduction, 32 do it sexually, 31 vegetative and 11 can do it both ways.

All this information has been useful in drawing up a guide for the sustainable management of invasive flora in Menorca.

# Invasive trend of *Parkinsonia aculeata* (*Fabaceae*) in Mediterranean Italy

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The development of intensive agriculture has modified the original structure of natural vegetation, allowing the direct or indirect introduction of exotic plant species, originally used in ornamental green areas or in reforestation but then escaped from cultivation. If for some species we can speak of a sporadic presence in nature, for others the invasive character is clear, characterised by a special ability to adapt to the territory and climate, often occupying biological niches already conquered by other species. Finally, for other taxa, it is only possible to know the potential risk of invasiveness on the basis of their ability to pervade the territory and their relative adaptability. The present work focuses on the distribution of *Parkinsonia aculeata* L. (*Fabaceae*) in Italy. It is a thorny shrub or small tree, drought-tolerant, often cultivated in parks and gardens in arid areas with a Mediterranean climate. The species is identified as an invasive neophyte, native to the Sonora and Chihuaha deserts of North and Central America where it is economically important for feeding small ruminants, and ecologically in fixing nitrogen in the soil. In Italy it is reported as a naturalised alien in Puglia, casual in Liguria, Calabria and Lazio, and invasive in Sardinia and Sicily.

Several species of this entity have obtained a certain autonomy in the coastal areas of Bari, with the presence of some spontaneous individuals on a slope of an alley road, and others in uncultivated land, in suburban areas of the territory of Bitonto (Bari), either by seed dispersal or by rooting from garden waste. In Calabria, the species is present with isolated individuals or in small units in the coastal area. In Sicily the first cases of naturalisation were reported as early as the 1970<sup>s</sup>, when the species colonised great coastal areas, especially along the Tyrrhenian coast, often together with the other hostile *Vachellia karoo* (Hayne) Banfi & Galasso of the same family *Fabaceae*, competing with native species, changing the structure of natural plant communities and the Mediterranean landscape.

## *Koelreuteria paniculata (Sapindaceae)*, casual alien tree in the South Caucasus and in the Mediterranean region

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The east-Asian species *Koelreuteria paniculata* Laxm. was introduced in Europe, in different parts with a temperate climate, in the year 1747. In South Caucasus it was first introduced in the area of today's National Botanical Garden of Georgia (Treasury Garden) in  $19^{th}$  century. The cultivation of artificial greeneries of *K. paniculata* in Georgia dates back to the  $50^{s}$  of the  $20^{th}$  century. In the Mediterranean region, especially in Italy, it is considered casual alien. In Sicily, it spread widely from the artificial greenery of the city to many uncultivated urban and peri-urban spaces. Its spread from artificial plantings is also noted in the South Caucasus, however there are no scientific data on its invasiveness.

To determine the level of invasiveness of *K. paniculata* in Georgia, we observed the vicinities of Tbilisi where it is cultivated in several locations. According to the preliminary observation *K. paniculata* shows good/satisfactory levels of vitality, however dead individuals were also present. The plantations are between 4-5 and 8-10 m height and the average diameter of the trees varies between 20-30 (35) cm. On every location *K. paniculata* grows via seeds and there are young and juvenile individuals of various ages. Shoots from the cut down tree individuals were also present. The regeneration of *K. paniculata* shows good results in less dense stands (density 0.3-0.5), while in relatively high density stands (0.7-0.8) its juvenile and young individuals are either absent or very small in numbers. The numbers of juvenile and young individuals are concentrated in artificial plantations and its edges. They do not separate (or slightly separate) from the artificial plantations and are not distributed within their neighboring natural grasses and shrubberies. However, naturalized individuals of *K. paniculata* were observed in various locations of Tbilisi area that are not associated with the artificial plantations.

The distribution of *K. paniculata* in the National Botanical Garden of Georgia must be mentioned separately, where it has well adapted and many different aged tree individuals are spread across almost the entire territory of the Botanical Garden. It is noteworthy that the level of invasiveness of *K. paniculata* within the Tbilisi area is significantly less compared to other introduced plants, such as: *Ailanthus altissima* (Mill.) Swingle, *Cercis siliquastrum* L., *Spartium junceum* L., etc.

**Poster Presentations** 

## Hydroalcoholic extract of wild *Sulla coronaria (Fabaceae)* flowers counteracts oxidative stress and inflammation in human dermal fibroblasts (HDF cells)

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The Fabaceae family includes the perennial herbaceous plant Sulla coronaria (L.) B.H. Choi & H. Ohashi, a species with origins in the Mediterranean basin who has been extensively cultivated for its agricultural and ecological advantages. The plant's ability to produce high-quality fodder gives it substantial farming importance. It is renowned for its capacity to fix atmospheric nitrogen, enriching the soil and obviating the need for nitrogen-based fertilizers. Its suitability for cultivation in a range of climates is further increased by the fact that it has good drought tolerance and can flourish in several soil types. Its deep root structure aids in soil stabilization, avoiding erosion and fostering land preservation. S. coronaria has been traditionally used in herbal medicine for its different medicinal properties. It is important to highlight that while S. coronaria has a history of traditional use for medicinal purposes, scientific evidence supporting its efficacy and safety is currently limited. Recently, it was reported that S. coronaria extract promotes collagen synthesis in vitro, suggesting that it is suitable for cosmeceutical products for skin disorders. Since numerous studies have demonstrated that natural compounds are valuable therapeutic agents targeting oxidative stress and inflammation in skin cells, in the present study, we evaluated the phytochemical profile and, for the first time, the antioxidant and anti-inflammatory inhibitory activities of S. coronaria flowers' hydroalcoholic extract collected Corleone (Italy), on HDF cell line.

The qualitative and quantitative analysis (HPLC-DAD, Folin-Ciocalteu, and aluminum chloride methods) of *S. coronaria* wild extract confirmed the good content of phenolic compounds (TPC 69.8  $\pm$  0.6 mg GAE/g extract, TFC 15.07 mg CE/g extract) and the significant presence of rutin, quercetin, and isorhamnetin derivatives. In addition, a good free radical scavenging activity was highlighted by the DPPH assay that showed an IC<sub>50</sub> of 8.04  $\pm$  0.5 µg/mL. Biological investigations showed the safety of wild *S. coronaria* extract and its capacity to counteract the inflammation state induced by Interleukin-1 $\beta$  exposure by reducing in a concentration-dependent manner the levels of both nitrites and nitrates (final metabolites of NO·) and ROS in HDF cells. In conclusion, our results suggest that the anti-inflammatory activity of *S. coronaria* extract is strictly associated with the antioxidant properties of the phytocomplex present in the flowers, and it may represent a valuable source of bioactive molecules for nutraceutical and cosmeceutical purposes. Its cultivation and utilization can contribute to sustainable agriculture practices, land conservation, and potential innovations in various fields of well-being.

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## Distribution, conservation and reproductive biology of the rare Greek endemic *Limonium corinthiacum (Plumbaginaceae*): an update

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*Limonium corinthiacum* (Boiss. & Heldr.) Kuntze is a Greek endemic, perennial, coastal plant species for which little is known regarding its biology and distribution. According to The Red Data Book of Rare and Threatened Plants of Greece the species is classified as Endangered, while the population on which its description was based, recognized by Heldreich in 1852, has not been found ever since. Apomixis, polyploidy, and hybridization are common reproductive mechanisms observed in *Limonium* populations, shaping the evolutionary history and current distribution of the genus in Greece. This study aims to add new data, and thus reevaluate the distribution, conservation status, and reproductive biology of *L. corinthiacum*.

For this purpose, *L. corinthiacum* populations were recorded during the summer months of 2020-2022, along the coastline of continental Greece, and specimens were collected for laboratory analysis. A distribution map was created using the coordinates of all known populations (field observation sites and data extracted from the literature) and the species was subsequently assessed according to the IUCN Red List Categories and Criteria. The research into the reproductive mechanisms involved seed production investigation, breeding systems' analysis, using differential staining of pollen grains, and an examination of the species' heteromorphic self-incompatibility, based on morphological differentiations of pollen grains and stigmas.

According to our study, *L. corinthiacum* shows a punctiform, discontinuous distribution, from the coastal area of Galaxidi town to the west, to the southeastern coasts of Attica, to the east of continental Greece. New populations of the species were discovered along the coasts of Corinthian Gulf and the southeastern coasts of Attica, while some old records were not confirmed, suggesting the species is becoming susceptible to a variety of threats, like some anthropogenic interventions and invasive species. However, based on the reevaluation of its conservation status, we propose downgrading *L. corinthiacum* to Vulnerable under B1ab(iii)+2ab(iii). Concerning reproduction biology, almost all analyzed populations exhibited finely reticulate pollen (type B) with low viability, papillate stigmas, and good seed production, a pattern that indicates apomixis as the primary reproduction mode. Finally, morphological and breeding systems' examination revealed a hybridization for the first time within *L. corinthiacum*, with the more common *L. aegaeum*.

To conclude, our study extended the distribution limits, revealed the reproduction mechanism, and provided new data for the conservation of *L. corinthiacum*, while also highlighting the necessity of fieldwork for rare plant species of the Greek flora.

## The genus *Limonium (Plumbaginaceae)* on Ikaria Island: taxonomy, distribution, ecology, and reproductive diversity

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*Limonium* Mill. (*Plumbaginaceae*) exhibits high levels of diversity in the Aegean Islands, a fact that is partly connected to the variety of its reproduction modes. Ikaria, an island of the East Aegean region, is noteworthy from a geological and phytogeographical point of view, and although quite well-explored, data regarding the presence and distribution of *Limonium* species on the island were still inconsistent.

In order to clarify the diversity of *Limonium* taxa growing on Ikaria and understand their ecological preferences, we conducted extensive fieldwork recording *Limonium* populations all over the island, identifying their geological preferences, and noting the altitude of their occurrence. Ample material was collected for taxonomic identification and reproductive systems` investigation. The geographical coordinates of the verified populations, as well as the locations where no individuals were present, were used to produce a distribution map for *Limonium* on Ikaria, using the ArcGIS Pro software.

According to our study, Ikaria hosts eight *Limonium* species, namely *L. atticum* Erben & Brullo, *L. dolihiense* Erben & Brullo, *L. graecum* (Poir.) Rech. f., *L. ikaricum* Erben & Brullo, *L. kirikosicum* Erben & Brullo, *L. proliferum* (D'Urv.) Erben & Brullo, *L. roridum* (Sm.) Brullo & Guarino, all of which are capable of reproduction via apomixis (either obligate or facultative apomicts). *L. sinuatum* (L.) Mill. is the only clearly sexual species on the island. *L. atticum* is reported for the first time in the phytogeographical region of the East Aegean Islands, whereas *L. proliferum* is a new record for the island of Ikaria. New localities are reported for all the remaining species. The plants were located at an altitude of 0 to 15 m a.s.l. The range-restricted taxa showed geological specialization, in contrast to widely distributed ones, that were recorded on a variety of substrates. Based on field observations, the abrupt uplift of the topographic background and the presence of extensive coastal areas consisting of boulders, incapable of any soil retention, form an unsuitable ecological niche for *Limonium* populations.

To conclude, the detailed mapping of *Limonium* populations and the study of their reproductive biology were conducted for the first time on the island of Ikaria. The observed high apomixis levels, combined with the geological heterogeneity of the island seem to favor numerous *Limonium* "microspecies" with restricted distributions. Similar studies on different Aegean islands may shed light on the distribution patterns of *Limonium* in relation to geological formations and facilitate the understanding of evolution and adaptation within the genus.

# Adaptive plasticity in regeneration traits of *Echium vulgare* (*Boraginaceae*) from contrasting habitats: a preliminary study

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Over time, studies of plants growing in contrasting environmental conditions were refined by considering the intraspecific functional trait variability (ITV) and related trade-offs, providing useful insights into species adaptive capacity. In nature, edaphically stressful habitats provide unique opportunities to study the factors and mechanisms promoting ITV. Among these habitats, the serpentine outcrops represent edaphic islands characterized by extreme physical-chemical anomalies: low Ca/Mg ratio, pH values ranging from basic to ultrabasic, nutrient deficiency, especially in nitrogen and phosphorus, phytotoxic concentrations of metals, dark soils subject to extreme heat and drought during summer. In such context, a previous study detected a relevant ITV in *Silene paradoxa* leaf traits, with the populations from serpentine sites showing traits polarised towards the stress-tolerant adaptive strategy.

In this preliminary study, we aimed to evaluate the intraspecific variability of the regenerative traits Seed Mass and Germination Rate in three populations of *Echium vulgare* growing across stressful and non-stressful environments. The fruits were collected from 75 and 30 individuals from, respectively, serpentine outcrops of Monte Ferrato (Italy; hereafter MFER) and Librazhd (Albania; hereafter ALB). Fruits were also collected from 50 plants on Monte Amiata (Italy; hereafter, MAMI) on non-serpentine soil. On average, the plants collected were 15 cm in height in MFER and 40 cm in MAMI and ALB. To evaluate variations in seed mass, 80 mericarps were randomly selected for each population (MFERR, ALB, MAMI), and weighed using an analytical balance. Germination rates were tested by placing ten replicates of 8 mericarps per provenance in seed germination boxes with filter paper moistened with deionized water. Germination boxes were placed in a growth chamber with a 12h/12h day-night cycle with a constant temperature of 20° C and 59% relative humidity. The duration of the germination test was 30 days, with a daily frequency of surveys. All the seedlings were transferred to pots with neutral soil and propagated for future surveys.

A non-parametric Kruskal Wallis test was performed to compare seed mass among populations, and showed significant differences between ALB and MFERR. MFER showed the lower seed mass values. No differences in seed mass were evidenced in relation to the native soil type. As for germination rate, soil type significantly affected the variability among populations, with the MAMI population clearly showing a lower percentage of germination and a later germination compared with the other two provenances, MFER and ALB.

### Mediterranean Medicinal Plants in old Georgian Medical Sources

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History of medicine in Georgia dates back to the ancient times. The medieval manuscripts of medicinal character (the so-called Karabadini, which is a common name for healing books in old Georgia) have survived in the archives until now. Part of these manuscripts have been studied and edited. Despite this the mentioned sources still contain a great number of unknown or unidentified medicinal plants. Among them can be found the species, which are not native for Georgia, on the one hand, and plants, known as the country endemics, on the other. It was also interesting to research the links between Mediterranean and Georgian medicine, and whether the medicinal plants of this region were in use in Georgia.

Search for the plants of Mediterranean origin in the old healing books was the aim of our research. In our study we used a new method of identification of plants mentioned in the manuscripts. According to this method similar prescriptions are searched in the medicinal manuscripts of different countries and by the comparison and identification of their components, and etymological analysis of local, folk names the before unknown plants are recognized.

For example, in the prescriptions given in 15th century Karabadini, authored by Zaza Panaskerteli Tsitsishvili, there was mentioned the root of the plant named "tiva katsi", which literally is translated as a "hay man". It was not clear, what kind of plant was meant under this name. When we compared the Georgian source with the work by Avicenna –" The Canon of Medicine", this unknown component fitted to the description of mandrake (Mandagora sp). If we look at the mandrake root, it indeed looks like a hairy man. This explains, why mandrake was called "tiva katsi". It is also interesting, that the root of "tiva katsi" is mentioned as well in the Georgian source Ustsoro Karabadini by Kananeli, dated by 10-11th centuries, which is older than Avicenna's book. The first mention of mandrake is met in Egyptian papyruses, the Bible and the ancient Greek medicinal sources. Mandrake is not native for Georgia - it was brought from the Mediterranean area and numerous legends are connected with it. It is worth to note that before our studies mandrake was not identified by the researchers in the healing books of Karabadini, which is a novelty of our research.

Using similar comparative approach, the presence of other mediterrannean plants, such as Greek lavender - *Lavandula stoechas* L ("ustekhudosi", "astovakhudosi") and cypress - *Cupressus sempervirens* L.was established in Georgian medicinal books Karabadini.

This fact is an additional proof of the links existing between Georgian and Greek medicines since ancient times. This is not surprising especially considering the old myth of Argonauts, where Colchis princess Medea was depicted as being experienced in the art of healing and the garden of plants with healing properties, referred to as "Medea's Garden" existed in the kingdom of Colchis.

# Recording the floristic diversity in archaeological sites in Greece: the sanctuary of Artemis in Vravrona, Attica

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Archaeological sites are often repositories for biodiversity conservation, since many human activities are restricted in these areas, thanks to their legal protection status. In the framework of an ongoing research project, the animal and plant diversity of twenty archaeological sites across Greece is being recorded. The project is implemented under an Agreement signed in August 2022 between the Ministry of Culture and Sports, the Ministry of Environment and Energy, the National and Kapodistrian University of Athens and the Natural Environment and Climate Change Agency. Among the twenty archaeological sites under study is the sanctuary of Artemis (Diana), the goddess of vegetation and hunting, in Vravrona, an important worship site in Antiquity. The present contribution is a preliminary presentation of the results of the floristic research at this site, which have been derived from a review of the relevant existing literature, as well as from fieldwork data. According to these preliminary data, the presence of 137 plant taxa (species and subspecies), belonging to 123 genera and 57 families, has been recorded. Among the most important findings are some Greek endemic taxa as well as some aquatic and hydrophilous plants, which are not particularly common in the relatively dry region of Attica. More specifically, the presence of the endemic Silene spinescens and Tragopogon longifolius and the relatively rare species in Attica, Alisma plantago-aquatica and Iris pseudacorus has been reported until now. In the above-mentioned number of plant taxa recorded so far, some old or more recent bibliographic references have been excluded, since these may concern the wider Vravrona area and not specifically the archaeological site. The field investigation will continue in the next few months until the completion of the project in the first half of 2024, aiming to record, as far as possible, the complete flora of the area.

### Ancient herbaria in Spanish schools

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Natural science studies have had great scientists who have published important papers throughout history. This knowledge has allowed us to discover the flora and vegetation of our territory. Some of these botanists are, and were, naturalists and religious people who were able to dedicate their free time to the recognition of the plants around them and, in some of the cases, they came to elaborate herbariums that have come down to our days. In many cases, it is in schools and institutes that these natural history collections can be found.

Many of these schools belong to religious orders like La Salle, Jesuits, Marists... In the second half of the 19th century, a new law made it compulsory for the institutes in Spain to have a natural history cabinet, and this would be the beginning of most of the museums that they still preserve nowadays. In 1901, France bans religious education and many of the religious brothers crossed the Pyrenees and settled in Spanish schools. That was when Frère Sennen (Étienne Marcellin Granié-Blanc; Coupiac 1861 – Marseille 1937) started to study the Iberian flora and prepared the exiccata "Plantes d'Espagne" and later "Plantes du Marco".

Some of the La Salle schools preserve herbariums collected by their brothers, who corresponded and sent each other specimens around the country, achieving extensive herbariums. A global review in Spain has allowed us to show the presence of some of these plant collections started at the second half of the19<sup>th</sup> century. Currently, it has been possible to verify about 150,000 specimens in 20 different schools.

Fortunately, most of these plant collections are in a good conservation state and only a small part of them have been studied. These show interesting herbarium sheets, as well as new chorological data or first mention of a taxon in Spain. Historical collections are also a part of the history of science, and we used them as a tool to biograph its collectors. However, most of these data remain outside the scientific scope due to lack of knowledge or involvement of the institutions.

### The Mediterranean world of Hans Sloane: insights from the SloaneLab

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The death of Sir Hans Sloane in 1753 led to the creation of Britain's first national collection. Bought by the British state on behalf of the nation, Sloane's collection was made freely available for "all curious and interested persons" through a new institution, the British Museum (BM). Sloane's vast collection included natural history specimens, books, manuscripts, prints, drawings, coins and other objects from across the world. As an apothecary and physician, botany was particularly important to Sloane, and his surviving botanical collections alone comprise an estimated 130,000 specimens from over 70 countries and territories worldwide. Assembled from c.1680 onwards, in part financed by profits from the transatlantic slave trade, Sloane's vast botanical collection was made as Britain became a global trading and imperial power. Anyone wanting to interrogate Sloane's collection today however, is hampered by its patchy digitisation and its distribution across three different institutions: the Natural History Museum (NHM), the British Library (BL), and BM. The SloaneLab project aims to address these issues, digitally unlocking data on Sloane's botanical collections. It is virtually reuniting Sloane's original collection catalogues and surviving specimens and objects from across the NHM, BM and BL, and supporting different ways to search those collections for any "curious or interested person". A unique and integral component of Sloane's botanical collection is the 'Vegetables and Vegetable Substances' collection. It originally consisted of 12,752 botanical specimens, most housed in small glass and wooden boxes. Sealed inside these boxes are parts of plants such as seeds, beans, leaves, bark and gum, that were collected between the 1680s and 1750s. Each object was numbered and a corresponding description of the item was entered into a manuscript catalogue. Today, more than 8800 specimens and the original three-volume manuscript catalogue survive. As part of the SloaneLab project, a transcription of the catalogue has now been made available and cross-referenced to surviving objects (https://data.nhm.ac.uk/dataset/sloane-vegetable-substances). The catalogue entries vary markedly and can include information about people and places and the local uses of plants; marginal annotations detail cabinet and drawer numbers where Sloane kept these things in his house, while bibliographic references and later determinations provide further insights into the material contained in the collection. This poster examines Sloane's 'Vegetables and Vegetable Substances' from the Mediterranean region. It considers the geographical scope of the collection, the range of contributors, the nature of the material Sloane received and why those objects are of interest today.

# Pine reforestation in Mediterranean mountains. An ecological problem or an opportunity?

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Conifer reforestation plantations in central Italy were massively made since the end of 1800 to the end of 1900. They were mainly made as forest restoration purpose, to foster the secondary succession (forced secondary succession), to stabilize slopes and to reassemble a hydrogeological balance.

In central Italian reforestation we can mainly find native species like *Pinus nigra* J.F.Arnold and *P. halepensis* Mill.

If these reforestations have been well carried out and well managed (i.e. they have not undergone cuts, cleanings, fires, etc.), they are working well in the Apennines; for this reason, we cannot promote campaigns for their eradication, also because there would be great damage and gaps for the mountains. We have to wait for these formations to slowly turn towards more natural stages; it is possible to experiment with thinning cuts which, however, do not have a particular function, given that these trees are easily knocked to the ground by natural events, such as snow, wind and ungulates, which prefer them to rub against them, consuming the bark at the base and causing death. We also must not believe that our mountains and ecosystems are so foreign to conifers; certainly, some species used are exotic, but on the other hand these could be areas destined for ex-situ protection of threatened species, furthermore the reforestation by native conifers could be considered as compensation following the destruction or the reduction of the original populations of those species.

As with the secondary formations, these conifers too can enter that circuit of areas to contribute to an eco-systemic and the landscape discontinuity, contributing to aesthetically embellish the landscape and enhancing their ecological value. Without considering that, in any case we could now speak of utility, historical-cultural peculiarities and beauty.

It is true that these formations are very subject to fires, but this does not mean that they should be mistreated and destroyed, given that other formations such as the Mediterranean macchia, garriga, oak woods, crops and secondary successions shrublands that are equally subject to this risk induced by man in our parts. Cutting and clearing these woods, like others, would be like choosing to drown the Mona Lisa in a cubic meter of concrete, just to preserve it from the ravages of time and vandals. Furthermore, we must not forget that often in some places these conifers are the only point that can give a certain illusion of wood and forest; they are better than nothing and certainly better than bare, eroding or parched land.

## *Lolium persicum (Poaceae)* discovered in Ancient Nicopolis, Greece, and the identity of the enigmatic *L. scholzii* revised

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Recent extensive fieldwork took place in twenty selected archaeological sites of Greece during 2023, aiming at recording their floristic and faunistic diversity. During a three-day visit to ancient Nicopolis near Preveza (Epirus, Western Greece), our attention was drawn to a particular annual member of *Lolium (Poaceae)*, prompting us to collect samples for further investigation and place them in the ATHU herbarium. By applying three different identification approaches the specimens keyed out as *Lolium persicum* Boiss. & Hohen. Identification was verified using the iconography provided by E.E. Terrell, the monographer of the genus, the Flora of North America Project, and herbarium specimens. *Lolium persicum* is thus a new record for the Greek flora.

The Nicopolis specimens of *Lolium persicum* do not fit the morphological variation of any of the seven known *Lolium* species found in Greece (*L. multiflorum* L., *L. perenne* L., *L. remotum* Schrank, *L. rigidum* Gaudin, *L. subulatum* Vis. and *L. temulentum* L.) except *L. scholzii* Greuter. The latter is a puzzling species collected only once, at the western foothills of Mt. Taigetos, south Greek mainland. Three herbarium sheets of this species are known (PAL, holotype; B and UPA, isotypes) of which the PAL and B specimens were consulted. *L. scholzii* fits *L. persicum* well on account of its slender annual habit, narrow, ca. 0.5 mm rachis, 3–5 florets per spikelet, and elliptic to lanceolate lemmas bearing a 3–7 mm long awn. When described, *L. scholzii* was compared with *L. rigidum* and this is apparently the reason why some authorities placed it, incorrectly, under the synonymy of the latter.

The recent discovery of *Lolium persicum* at the ancient Nicopolis poses a question regarding its origin. With a history dating back 2,000 years, as the capital city of the Roman province of Epirus Vetus, Actia Nicopolis offers a range of possibilities on when the species might have been introduced. On the other hand, *L. persicum* is indigenous along the coasts of Anatolia, i.e., close to the border of Greece with Turkey; therefore, its presence in Greece as a native species cannot be ruled out. The behavior of *L. persicum* as a weed of cereal fields is responsible for its introduction to several distant parts of the world, like Belgium, Canada, the USA and China. Thus, it may have reached Nicopolis as a weed of arable land, although the specimens collected in 2023 were not related to any form of cultivation.

# The Costa Brava as a focus of plant invasions: chorological novelties resulting from the first steps of the LIFE medCLIFFS project

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The Costa Brava, the coastal strip of ca. 250 km stretching from the French-Spanish border to Blanes, is known for its rugged appearance, with abundant sea cliffs, in clear contrast to other nearby coastal areas characterized by long sandy beaches. As a result of the interaction of various factors, including a complex topography, a profound transformation of the habitat and enormous tourism development, the Costa Brava has become a focus of plant invasions, probably the most important in the entire Iberian Peninsula.

In order to prevent new introductions of alien plants and improve the management of invasive species already present, a project within the LIFE Program of the European Union is underway. The initial phase of this project (LIFE medCLIFFS; https://lifemedcliffs.org/es/) required the design of a set of transects (around a hundred) of 0.5–1 km in length distributed throughout the coast of the Costa Brava; these small strips of coastline are being monitored by volunteers with the aim of early detection of new invasive plants and monitoring the different species already observed.

As a result of the fieldtrips derived from the design of the transects, which were carried out during the first half of 2022, we were able to detect a large number of species observed as subspontaneous for the first time in this coastal region, some of which are novelties even at a European scale. We also provide updated information on other invasive species that were already present on the Costa Brava but for which our observations significantly change the knowledge about their distribution range and invasion status in this area.

# The evolutionary history of junipers (*Juniperus* sect. *Juniperus*) based on genomic (GBS) analysis

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Junipers are trees or bushes belonging to the cypress family and distributed across the Northern Hemisphere. Depending on the taxonomic treatment, Juniperus is recognized to encompass 53 to 76 species, of which 10 to 14 are included into J. sect. Juniperus. Previous phylogenetic studies support the idea that J. sect. Juniperus constitutes a natural evolutionary group, which includes plants restricted to the Old World region (with the exception of the circumboreal distributed J. communis). However, little is know about the phylogenetic relationships within the section due to the low resolution of the genetics markers used up to date. To resolve this puzzle, we presented a reduce-representation genome (GBS) strategy including samples from all the species considered in J. sect. Juniperus. To compare the performance of different assembly methodologies, we run both denovo and reference (using Cupressus sempervirens genome as reference) assemblies testing several parameters in order to get the most informative alignments. The referenced dataset includes a higher number of Pis (parsimony informative sites) and recovered loci, but it also presents the highest percentage of missing data. The phylogenetic inference methodologies used for tree reconstruction, based on either a concatenated supermatrix or summary gene tree methods, were performed for both assemblies strategies (reference or denovo). The results concludes that J. sect. Juniperus comprises two natural lineages: one including species related to the J. communis complex, which includes the Far East distributed representatives, and another group including J. oxycedrus (prickly juniper) and allies, with a narrower distribution encompassing the Mediterranean area and part of the Macaronesian archipelagos. We discuss the taxonomic status of various members of the last lineage, confirming the taxonomic species-level range of several taxa traditionally recognized as subordinate to J. oxycedrus. These taxa include J. badia, J. deltoides, J. cedrus, J. maderensis, J. navicularis and J. willkommii. Additionally, we present a dated phylogeny to track diversification rates within J. sect. Juniperus.

# Exploring the evolution and speciation drivers of rock-dwelling endemics in the genus *Daphne* (*Thymelaeaceae*) within the Alps and Balkans

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Rocky habitats, which serve as ecological islands within diverse landscapes, exhibit remarkable biodiversity across various organismal groups. In Europe, concentrated primarily within the middle altitudinal zones of mountainous regions, these habitats provide a home to a diverse array of endemic species. One genus that typically inhabits rocky habitats is Daphne L., which encompasses deciduous or evergreen shrubs native to Europe and Asia. Approximately twenty European Daphne species are endemics restricted to specific mountain ranges or islands within the European Alpine System, with the Mediterranean region hosting the highest concentration of them. To deepen our understanding of the evolutionary processes and diversification of species endemic to rocky habitats, our project focuses on taxa found in the Southern and Southeastern Alps, as well as the Balkan region. Despite the recognition of many of them as prominent endemics within their respective regions, a significant knowledge gap persists regarding various aspects of their biology and ecology. Our research endeavors to bridge this gap by investigating the morphological, karyological, and genetic diversity of these species. Additionally, we aim to unravel the mutual interactions between these species, (hybridization and introgression), and uncover their role as speciation mechanism in the genus. We will also explore crucial facets of their reproductive systems, including the efficacy of sexual reproduction, pollination mechanisms, seed biology, and the extent of vegetative reproduction. Last but not least, we will address the role of root mycorrhizal fungal assemblages in facilitating the adaptation and survival of these species within rocky habitats. Through our comprehensive approach, we seek to shed light on the evolutionary and speciation processes that have shaped the rock-dwelling endemics in mounbtauns of European Alpine Sytem.

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# Ecological and morphometric study of the genus *Carpobrotus (Aizoaceae)* in the Western Mediterranean Basin

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The genus *Carpobrotus*, which belongs to the family *Aizoaceae*, has more than a dozen species that are native to the Southern Hemisphere (Oceania, South America and, more importantly, Southern Africa). However, several of these species have naturalized outside their native range as a result of being used as ornamental plants, as well as for stabilization of coastal dunes. The genus has become invasive in many parts of the world, posing a great threat to biodiversity. As of today, there are three species within this genus that are considered invasive, *Carpobrotus chilensis* (Molina) N. E. Br., *C. edulis* (L.) N. E. Br. and *C. acinaciformis* (L.) L. Bolus, which are known to produce hybrid forms and result in a taxonomic complex that is hard to tell apart from the parental forms. Therefore, the taxonomy and biogeography of the genus *Carpobrotus* remains unclear and under discussion.

The Western Mediterranean Basin is home of numerous populations of this genus, that, despite being initially assigned to *Carpobrotus edulis* for most of the cases, recent studies suggest that they could belong to different species and, probably, to the hybrid forms between them. In this study we measured several morphological characteristics of the reproductive and vegetative organs from over 30 populations stretching from southern Spain to southern Italy. We also extracted the bioclimatic variables from a much wider set of populations (nearly 400) from the same geographic area. These data were used to (i) test whether different entities can be identified based on morphometric characters, and (ii) if yes, to search for differences in their ecological niche.

### The decline of the native *Xanthium strumarium* and the invasiveness of the American *X. orientale*. Are competition and allelopathy triggering factors?

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*Xanthium* L.is a widespread genus of the sunflowers family (*Asteraceae*). In Europe we can recognize three species. *X. strumarium* L., *X. orientale* L. and *X. spinosum* L.

The former two are part of the same section and are similar in both morphology and ecological requirements and are often confused in the field. *Xanthium strumarium* is native of Europe, whereas *X. orientale* originated from America and has become widespread due to human mediated dispersal. They can be distinguished through the morphological features of the female capitula (burs), which are smaller and with less spines in *X. strumarium*, bigger and fully covered by spines, in *X. orientale*.

The American species has been observed in Italy starting from 1820 and has spread within a century to the whole Italian territory and most of the Mediterranean basin. In the meantime, *X. strumarium* has experienced a significant decline, but the specific reasons behind it remain unclear.

The genus *Xanthium* comprises species capable of synthesizing allelopathic compounds with inhibitory effects on surrounding plant species.

Therefore, we wanted to investigate if the presence of *X. orientale* inhibits the germination and growth of the European congeneric through allelopathy. For this purpose, we conducted germination experiments to determine the potential allelopathic effects of exudates obtained from dried leaves of *X. orientale* on the germination and growth of *X. strumarium* seeds. Moreover, we carried out competition trials to test the effect of the interspecific competition of *X. orientale* over the native species.

We compared the germination rate, germination time (days after sowing), and growth of the seedlings of the two species under the treatment (watering with *X. orientale* exudates) and the control conditions. The presence of a significant effect has been observed both on germination rate and growth of the two species, while the germination time was affected only at intraspecific level (*X. orientale* over itself) and not in *X. strumarium*.

The measures of fitness chosen for the competition trials were dried biomass, number of burs and bur biomass. ANOVA analyses have shown a significant effect of the interspecific competition of the invasive species over *X. strumarium*. An intraspecific competition effect was observed albeit to a much lesser extent.

# Identification and genetic characterization of centenary olive trees in the Sicani Mts (CW-Sicily)

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The olive tree (Olea europaea L.) is a long-lived tree that characterizes Mediterranean environments, appreciated for its fruits and oil. The history of the olive tree is intertwined with that of the populations of the Mediterranean basin. The cultivation of the olive tree in Sicily has very ancient origins and a very rich varietal heritage characterizes the island which, nationally and internationally, stands out in the production of strongly typified extra virgin olive oils. Furthermore, many monumental, little characterized, still productive, olive trees enrich the territory. The importance of protecting this heritage, which has long remained unknown even to those who work in the sector, is determined by the commercial interest in the "diversity of oils" and as a genetic source of resilience to abiotic and biotic stress. Added to this is the risk of genetic erosion due to land abandonment that makes the traditional olive groves neglected and more vulnerable to the fires, leading to the destruction of many centuries-old olive groves located into the Sicani Mountains. In this work, the monumental olive trees of the "Monti Sicani" area have been studied for the first time at a molecular level; these are both plants grafted on wild olive trees and ungrafted. The analysis was conducted using a set of 9 microsatellite markers, including the genetic profile of a set of standard cultivars and other monumental trees as a control. A cluster analysis was also performed which highlighted the genetic variability of some phenotypes typical of the area under examination. The UPGMA dendrogram further highlighted the proximity of some clusters of 'Nocellara' genotypes to the cv. Nocellara del Belice and instead a certain discrimination of 'Biancolilla centinara' (BCC) with respect to the standard cv. 'Biancolilla'. Furthermore, ten trees were found to have quite distinct profiles, representing a new source of diversity. The SEL tree grouped with a centenary oleaster tree "Oleastro Inveges" (Sciacca, Sicily). This recovery and identification action is important to deepen the knowledge of the olive heritage to preserve monumental plants, often unique and at risk of genetic erosion, which can be useful for future evaluation and breeding programs. The results obtained will be compared with the morphological characteristics of the standard cultivars taken into consideration to compare them also from a phenotypic point of view in order to highlight their respective pomological diversity and to be able to discriminate them from the regional and national olive heritage.

#### The genus Aloe (Asphodelaceae) in Sicily

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The genus *Aloe* includes about 630 species mainly native to tropical and southern Africa, Madagascar, Middle East, and various islands in the Indian Ocean. A few species have also become naturalized in the Mediterranean area, India, Australia, North and South America, Hawaiian Islands, etc.

Vegetative reproduction is prevalent in the genus and many of the species are cultivated in the gardens and homes as ornamentals and a few species for Medicinal purposes.

In Sicily there are 13 species commonly cultivated and 6 of them are reported as casual or naturalized: *Aloe arborescens* Mill., *A. brachystachys* Baker, *A. maculata* All. subsp. *maculata*, *A. perfoliata* L., *A. vera* (L.) Burm.f., *A. × caesia* Salm-Dyck. These species entered the Sicilian territory as cultivated plants and were then involuntarily spread throughout the territory, often with garden cleaning residues that are thrown into uncultivated land. Their characteristics of resistance to aridity, fast growth rate and lack of palatability for herbivores allow their diffusion along the coastal areas of the island.

*Aloe vera* is the first species that was cultivated and became naturalized in Sicily. It seems that its cultivation on the island, as well as in many other parts of the Mediterranean, is due to the Arabs who spread this species for its medicinal properties. In fact, it is found on the carbonate reliefs of the Tyrrhenian coast near inhabited centers such as La Rocca of Cefalù and the promontory of Termini Imerese.

Over time, other species have come into cultivation, the large part as ornamental. The taxonomic study of the genus *Aloe* in Sicily is testified by the numerous taxa that have been described starting from the plants cultivated in the Botanical Garden of Palermo by Agostino Todaro and Antonino Borzi.

*Aloe arborescens* is the other species cultivated both as ornamental and for medicinal purposes. This species is still used today for the production of aloe gel and preferred to *A. vera* due to its greater development and higher growth rate. It is currently widely cultivated in the territories of Custonaci (TP), Cattolica Eraclea (AG), Paternò (CT) and Lentini (SR), and new fields are planted every year.

#### Seed viability and ex-situ conservation of Staphylea colchica (Staphyleaceae)

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*Staphylea colchica* Steven (*Staphyleaceae*), Caucasian bladdernut, Kolkhuri jonjoli in Georgian - is a deciduous shrub or a small tree (2–4.5 m tall), included in the Red List of Georgia (2014) with status VU, based on the IUCN criteria A2d;B1b (small fragmented distribution range).

The species is naturally distributed in the deciduous woodlands in the west and central parts of Georgia at altitudes up to 1400 m ASL, where it grows as an understorey plant with other woody species: *Castanea sativa*, *Quercus iberica*, *Fraxinus orientalis*, *Acer campestre*, *Buxus colchica*, *Hedera colchica*, (Terjola population, Accessions #CRSB:292 (2006), #CRSB:1637 (2019)), Corylus avellana, Sambucus ebulus (Khoni population, #CRSB-WT-54 (2017)).

Sweet scented and aromatic inflorescences of *Staphylea colchica* are intensively collected in forests for the preparation of the pickled side dish, traditional for the Georgian cuisine. The species is cultivated as an ornamental due to its attractive habit, fragrant inflorescences and papery seed pods, and as a melliferous plant.

Intensive collecting of inflorescences for food purposes puts natural populations of the species under pressure.

Structural peculiarities of seed formation, potential and actual capacities for the seed set, viability and germination capacity of freshly harvested seeds of *Staphylea colchica* and those stored at -20° C in the National Seed Bank of Georgia, National Botanical Garden of Georgia (NBGG), have been studied. The oldest examined seed collection has been kept for 17 years and the two other seed accessions were deposited in 2017 and 2019 (all collections are collected and duplicated to the Millennium seed bank of the Royal Botanic Gardens, Kew under the projects "Saving the Flora of Caucasus" (2010–2020) and "Garfield Weston Global Tree Seed Project" (2015–2019).

Germination and viability tests were carried out under laboratory conditions.

Collection of documented 5–6 years old seedlings, obtained from the seed, harvested in places of natural occurrence of *Staphylea colchica* is established on the experimental plot of the Department of Plant Conservation of the NBGG. Freshly harvested seeds sown in pots in soil substrate in 2016 germinated on the 5th month. Germination of seeds made 55%. Part of saplings after two years was transferred to the open ground.

Studies of the seed formation process in adult trees grown at the Plant Conservation Plot of NBGG revealed that a small number of seeds – on average 3 – is developed in fruits of *Staphylea colchica*. The produced seed is of good quality and is characterized by the well-developed dicotyledonous embryo and endosperm. Reasons for the low seed production capacity are discussed. The results are compared with the reports of some authors, who suggest that propagation of *Staphylea colchica* by seed is limited in the wild populations as well.

Results of this study and conservation work are important for the clarification of seed storage behavior of *Staphylea colchica* and development its germination protocol.

# Dunal and riparian vegetation in the Oriented Natural Reserve "*Laghetti di Marinello*" (Sicily): a highly dynamic ecosystem

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The Oriented Natural Reserve "Laghetti di Marinello", north-eastern Sicily, includes a coastal brackish system beneath the promontory of "Capo Tindari".

Starting from the last decades of the 19th century, literature reports the existence of the brackish system, which reached the current structure in the 20th century. The whole lagoon is in continuous transformation due to the extreme dynamics of the delimiting sandy bars. On this unstable environment, the vegetation is characterized by rapid migrations following the dunal shift.

The riparian vegetation appeared almost stable in the last decade, with the typical freshwater association *Phragmito australis-Magnocaricetea elatae* Klika facing the promontory, and the euryhaline association *Juncetea maritimi* Br.-Bl. facing the sea.

In the sandy areas surrounding the ponds, the *Hyparrhenion hirtae* Br.-Bl., P. Silva et Rozeira association, a predominantly thermophilic herbaceous and not purely psammophilous vegetation, was observed. Within the latter one, the exotic *Cenchrus setaceus* (Forssk.) Morrone, since over ten years formed a large population on the long-established sandy stretch.

Noteworthy, numerous young plantlets of *Pinus halepensis* Mill. occurred, invading a large part of the *Hyparrhenia hirta* (L.) Stapf grassland, foreseeing the onset of a thermophilic forest.

In conclusion, while the geological-structural characteristics of Laghetti di Marinello and the modeling action of marine and atmospheric agents have shaped a unique landscape that yielded the site worthy of protection and conservation, no less importance should be paid to the floristic aspect, whose peculiarities deserve specific valorization actions from an environmental, historical-cultural and didactic-scientific point of view.

#### *Securigera varia (Fabaceae)* in the Nebrodi Mountains, new for Sicilian flora

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According to the literature, *Securigera varia* (L.) Lassen [Bas. *Coronilla varia* L.] in Italy is present in all the peninsular regions; it is absent on the two major islands, Sardinia and Sicily. Recent floristic surveys on the Nebrodi mountains allowed to find it on the outskirts of San Fratello village (Messina province, N-E Sicily). The small population, made up of about ten individuals, is settled at the base of an escarpment, upstream and on the edge of the provincial road which leads to Cesarò. To date, there is no data on the presence of other groups of the species in nearby locations. Therefore, on the basis of current knowledge, the Nebrodi territory find constitutes the first report of the presence on the island of the unmistakable species whose provenance, due to its location on the outskirts of a high hill town (about 775 m a.s.l.), could be referred to a southern geographical transgression favored by zootechnical activities, which are very widespread in the territory. In the light of this, until further data are available, the reported Sicilian record must be considered as the causal presence of the species outside its natural range. In any case, the species's population dynamics in the first location, as well as any expansion in other sites, should be monitored over time.

*Exsiccatum*: Sicily, Nebrodi Mountains, clay soil, ca. 750 m asl, 37°59'59.13"N, 14°36'11.03"E, 20.06.2023, *Giarratana* et *Raimondo* (PAL, PAL-Gr, FI).

#### Plants and their distribution in the Balio Garden of Erice (N-W Sicily)\*

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The results of the survey of the florula established in the historic garden of Balio (Erice) and the mapping of the phytoindividuals of the species hosted therein are presented. This survey allowed to have a complete picture of the taxa present and of their spacial distribution and was aimed at drafting the project of restoration and enhancement of the same garden. The decorative florula of the Balio consists of 70 specific and infraspecific taxa; 56 genera referring to 36 families are represented. The presence of lauriphylls and evergreen sclerophylls, mainly trees and shrubs autochthonous with Mediterranean-Atlantic climate requirements, is a remarkable character of the Erice garden. The survey has also the presence of rather unusual or barely represented trees in Sicilian gardens, such as Taxus baccata L. and other Mediterranean-Atlantic elements including Laurus nobilis L. and Buxus balearica Lam. The frequency of the latter taxon is a clare demonstration of the climatic pecularieties of the place to host elements with oceanic climate requirements. Among the plants that contribute to giving a Mediterranean imprint to the garden, there are almost all the species present in the woods of the surrounding hills as Quercus ilex L. and Fraxinus ornus L., F. angustifolia Willd. subsp. angustifolia. Abundant - in many cases invasive - is Hedera helix L., but also Laurus nobilis. Among the shrubs, Chamaerops humilis L., Pistacia lentiscus L. and Viburnum tinus L. occur frequently. An interesting fact emerged from the study helps at understanding the ornamental flora present in the garden. In fact, the presence of some native species typical of the surrounding area has been underlined. They are perfectly integrated with the rest of the existing system. Apart from the aforementioned woody species, occurring the bush of Silene fruticosa L., Dianthus rupicola Biv. subsp. rupicola, Lomelosia cretica (L.) Greuter & Burdet, Athamanta sicula L., and Centaurea erycina Raimondo & Bancheva; transgressive, in the same context, is the endemic Brassica villosa subsp. drepanensis (Caruel) Raimondo & Mazzola. Among the annual plants, the site hosts the endemic Silene nefelites C. Brullo, Brullo, Giusso & Ilardi. Rich is the epiphytic florula, index of high atmospheric humidity. It essentially consists of bryophytes and both crustose and leafy lichens. Of the first contingent we recall the rare and significant presence of Cryphaea heteromalla Hedw., of the second the characteristic Usnea barbata (L.) F.H. Wigg. The analysis of the decorative florula of the garden, surveyed inside and in the immediate vicinity, together with the map of the phytoindividuals produced, constitute the basic elements for the development of the restoration and enhancement project: At the edge of the area, it provides an exemplification of the phytodiversity garden, with the typical elements of the Mediterranean maquis and garrigue, in addition to the main rocky species including the mentioned local endemics. In conclusion, the Balio historic Garden is characterized by a close relationship between cultural and natural elements of the place. In it, garden, rock, medieval architecture and natural vegetation are harmoniously integrated and this produces singular scenic effects.

\*Results of the botanical investigation carried out prior to the executive planning of the restoration and enhancement of the Balio historic garden, subject to a specific loan granted by the Ministry of Culture of the Italian Government, with funds from the PNRR.

## *Vachellia karroo (Fabaceae)*, alien naturalized and invasive in Sicily: its influence on natural ecosystems and the Mediterranean landscape

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In recent years, some woody *Fabaceae*, previously present sporadically in the Sicilian territory, have gradually spread to the point of visually influencing the landscape of the urban suburbs and even in some natural environments, such as those of the Mediterranean maquis and torrents, mainly near the coasts. In addition to *Robinia pseudoacacia* L., whose spontaneous diffusion is limited to some hilly and mountainous sectors of the territory of Messina, this contingent of woody taxa consists of *Acacia saligna* (Labill.) H.L. Wendl., *Leucaena leucocephala* subsp. *glabrata* (Rose) Zárate, *Parkinsonia aculeata* L., *Senigalia visco* (Lorentz ex Griseb.) Seigler & Ebinger, and *Vachellia karroo* (Hayne) Banfi & Galasso. *Vachellia karroo* and *Acacia saligna* are now elements of landscape expressiveness along the northern, southern and eastern coast of the island, especially during the abundant and continuous flowering that affects from spring to summer. The other taxa are more or less localized with groups scattered in the territory.

In addition to the entire coastal belt, in particular that of the western part of the island, *V. karroo* is also present in the interior hills up to an altitude of 750 m a.s.l., such as in the territories of Corleone and Monreale in the province of Palermo and in Bronte (province of Catania). In Sicily, the species bears fruit abundantly and easily regenerates. In all environments, even nitrophilous, *V. karroo* competes with other woody exotic species such as *Parkinsonia aculeata* and *Acacia saligna*. In addition to the main island, it is naturalized in most of the circum-Sicilian islands (Lampedusa, Pantelleria, Favignana, Levanzo, Ustica and part of the Aeolian Islands).

Due to the invasiveness perceived with progressive intensity in the last 20 years, in particular in the western provinces of Sicily, the wide diffusion of *V. karroo* – initially introduced to fix the dunes of the southern coast and then planted here and there to create hedges with trees border of private properties, due to the thorniness of its branches – can be connected to the ongoing climate change. It, therefore, constitutes a naturalized element to be monitored, controlled and with which to prudently coexist.

### Lectotyping of *Arthopyrenia parolinii* Beltr. (*Ascomycota, Trypetheliaceae*): investigations about a lost lichen species

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As part of the research focusing on the genus *Arthopyrenia* in Italy, we aimed to establish the identity of *Arthopyrenia parolinii* Beltr., a neglected species not yet typified.

This study is based on analysis of the protologue, pinpointing the location of the *locus classicus*, search for and examination of the original material.

In the protologue, Beltramini provides a diagnosis, the description, and drawings showing microscopic characters. The epithet is in honor of the "Cavaliere Nob. Alberto Parolini", an Italian naturalist known for his rich botanical garden (Parolini Garden) in Bassano del Grappa (Vicenza, Italy). The collecting site is mentioned as "*le tiglie nel passeggio di Belvedere in Bassano*" [on Tilia along Belvedere stroll in Bassano]. In Bassano del Grappa, the avenue once known as *Viale Belvedere* is now named as *Viale delle Fosse*. This avenue was built in 1790. After this work, a double row of linden trees was added to the avenue which reached the Parolini Garden. This new tree-lined avenue was called "*Passeggio pubblico di Belvedere o Fosse*" (Public walk of Belvedere or Fosse), the same name reported in the protologue.

The sample stored in the Lichenological Herbarium of A.B. Massalongo in VER is made up of a piece of bark glued to the herbarium sheet. The label reports the location "Tilia di Bassano" and the note "Herb. Beltramini". A calligraphic comparison made with original material, allowed us to identify the location as written by Beltramini while the name of the species and the herbarium as written by Massalongo. We also found a second undated collection of *A. parolinii* in M. This sample shows the same set-up. On the sheet, notes written by Massalongo are reported: the name of the species accredited to Beltramini, the note "nov. spec ?", the substratum "*ad Tiliae truncos*", the location "*Pr Vicez*." [Province of Vicenza], the herbarium from which the sample was taken ("*herb. Massalongo*"). The last specimen – under the name *Spermatodium parolinii* (Beltr.) Trevis. – is included among Trevisan's exsiccata deposited at MSNVE. Considering the stormy relations between Trevisan and Massalongo and the absence of notes on the specimen, it is conceivable that the collection was carried out by Trevisan himself and it should be not original material. Consequently, among the few original materials that currently exist, we designate the specimen stored in VER as lectotype, as this is the most complete, informative, and in line with the protologue.

# *Isoëtes delilei (Isoëtaceae)* in the temporary ponds of the Mamorà forest (N-W Morocco)

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An article by Medjahdi & Letreuch-Belarouci [Fl. Medit. 33: 61-65. 2023] reports for the first time in Algeria *Isoetes delilei* Rothm., a species included for a long time in *I. velata* A. Br. (= *I. longissima* Bory). It is therefore a taxon with a controversial history clarified recently with a clear separation from *I. velata*.

*I. delilei* is a small pteridophyte of the western Mediterranean, present in France, Spain, Portugal and recently also reported in Morocco, in the regions of Sidi-Bettache, between Casablanca and Rabat, and Jebilet (Marrakech). The analysis of the Algerian finding has allowed us to identify an extensive population of *I. delilei* observed and collected a few years ago at the margins of ponds and ephemeral pools recurring within what remains of the Quercus suber forest of the Mamorà, near Rabat (Morocco), today partly occupied by agriculture, partly also urbanized. Like most of the species of the genus, I. delilei is an amphibious plant, with a very small underground stem of an almost rectangular shape, devoid of phylloids; it has 9-18 thin and fragile membranous leaves, 10-25 cm long, of a glaucous green color and dilated at the base. The sporangia are naked, not covered by a veil, which clearly distinguishes it from *I. longissima*. The macrospores are sub-spherical and have protruding angular ribs. As also in the rest of the North African countries, the preferred habitats are very limited and highly endangered as they are occupied by agriculture. These are flat and humid lands affected today by intensive forms of agriculture or overgrazing. The record from Mamorà forest is very considerable but are exposed to the same dangers. In the surveyed environments the species lives together with Mentha pulegium L., Spergularia bocconei (Scheele) Grabn., Ranunculus muricatus L., R. trilobus Desf., Juncus bufonius L., Poa annua L. and other ephemeral microtherophytes. The species is everywhere considered threatened with a threat status ranging from NT (Near threatened - species close to the threshold of threatened species or which could be threatened if specific conservation measures were not taken, in Europe and in the world ) and VU (vulnerable) in France). In the Maghreb I. delilei with the stations of Morocco and the station of western Algeria, is critically endangered (CR). As has been underlined several times, urgent measures are necessary for the conservation of the Mamorà forest in order to protect Isoëtes delilei together with the internal humid habitats and therefore also.

#### Sex ratio shifts in the moss genus *Ceratodon*: implications for environmental adaptation in the populations of the south of the Iberian Peninsula

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This study aimed to investigate the hypothesis that the sex ratio of the moss species *Ceratodon purpureus* and *C. amazonum* populations in the southern areas of the Iberian Peninsula is significantly lower due to increasingly unfavorable environmental conditions. Also, we wanted to explore the potential consequences of these sex ratio shifts for the species' genetic variability and determine if male individuals are more adversely affected than females as we move toward the south. We collected 294 samples of *Ceratodon* from 67 localities across central and southern parts of Spain, selected to represent a range of environmental conditions prevalent in the study area. Given the morphological challenges in distinguishing male plants and the common absence of anteridia, we used two methods to detect male plants: the presence of sporophytes and the sequencing of the sex-linked molecular marker known as rpS15A in not sex-expressing plants (in 51 *C. purpureus* and 27 *C. amazonum* randomly taken samples).

Among the C. *purpureus* samples, 15 were fructified, and 49 of the sequenced plants were identified as females, with only two males detected. In the *C. amazonum* samples, only one was fructified and 26 out 27 individuals were females, with one male detected. These data indicate an obvious predominance of females in both species and reveal a remarkable sex ratio skew of their populations. Notably, a single population located in the far south exhibited the presence of both male and female *C. amazonum* samples. Investigating the specific environmental conditions of this site could offer valuable insights into the interplay between the environment and sex ratio. However, the statistical correlation analysis was constrained by the scarcity of male samples in other populations but provide initial insights into the potential influence of mixed environmental factors on sex ratio.

In conclusion, this study provides evidence of sex ratio shifts in *C. purpureus* and *C. amazonum* populations in the southern Iberian Peninsula. The results support the hypothesis that the environment may influences sex expression, potentially impacting the genetic variability and reproductive strategies of these species. Further research with larger sample sizes and extended geographic range is essential to deepen our understanding of the mechanisms driving sex ratio shifts and their ecological implications amidst ongoing climate change.

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# Genetic diversity and fruit variability of *Olea europaea* var. *sylvestris* (*Oleaceae*) in Menorca (Balearic Islands, Spain)

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Wild olive tree (*Olea europaea* var. *sylvestris* (Mill.) Rouy, known locally as "ullastre") is the most abundant and frequent tree in Menorca. It is the tree that characterizes the vegetal landscape of the island. It is the main component of the woodland islets that give character to the local mosaic landscape, it also forms extensive wooded masses, known locally as "ullastrars", which are considered a specific plant community: *Prasio-Oleetum sylvestris*.

Wild olive tree is economically important worldwide for being the predecessor of the olive tree (*O. europaea* L. var. *europaea*), in Menorca it is also an important resource in raw materials such as wood. In the island it is easy to observe how there is great variability in the shape, size and ripening time of the fruits, to the point that it is often not easy to establish a clear difference between wild plants and the olive cultivars that produce a small fruit.

To determine the economic potential of wild olive trees, a genetic and morphological study was carried out with three main objectives:

- i. Knowing the genetic diversity in Menorca;
- ii. Determine if any genetic group or race may be of interest for oil production;

iii. Find out if there is any correlation between fruit morphology and genetic diversity.

Plant material (leaves) and fruits were collected from 28 different localities, distributed throughout the island. The genetic analysis of 116 individuals resulted in the existence of 6 genetic groups. Group IV was the most represented with 65 individuals (57%), the second was group II with 29 individuals (26%), the other four had a representation of 5% or less.

The morphological study analysed the length (L), the width (W) of the fruit and the L/W ratio. The main result was the extreme variability of the fruits, from less than 9 mm to more than 15 mm in length, from completely spherical to narrowly elliptical.

The main conclusions of the study were:

- i. In Menorca, the genetic diversity of wild olive trees does not have a geographical pattern;
- ii. Nor is there any geographical pattern in the morphology and dimensions of the fruits.iii. There is a group with large fruits, most of them in genetic group IV;
- iv. It is possible to visually differentiate large fruit plants with more economic potential;
- v. The systematic cultivation of large-fruited wild olive trees would transform them into olive cultivars.

#### A botanical garden committed to attracting young visitors, improving their experience and encouraging scientific interest

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Marimurtra is a Botanical Garden located in an exceptional location, with a very favourable climate on the emblematic cliffs of the Costa Brava. It has a peculiar origin since its creation was the dream of one person, Carl Faust, a German businessman with a great sensitivity for nature and science, who surrounded himself with great botanists (such as Pius Font i Quer and Sventenius) to create a garden where scientists could study plants from all over the world.

In 1951, one year before Carl Faust passed away, he created a foundation named after him, Fundació Privada Carl Faust, which to this day continues to pursue objectives such as the conservation of plant species, research and scientific dissemination of the plant world. Today, the Marimurtra Botanical Garden receives a large number of visitors each year from all over the world, with a wide range of backgrounds, ages and interests.

Over the last few years, Marimurtra has been working to make its educational activities, especially those aimed at younger audiences, more experimental and manipulative. In short, seeking to generate more experiential experiences that really encourage interest in botany and curiosity about the environment. All of this, avoiding lectures and adapting the content to each educational stage.

Apart from having made a change in the format of educational activities, Marimurtra is working to attract this type of public, increasing the number of visitors in the scholar stage. This is a target audience and is of great interest to the foundation, which aims to have a positive impact on this group.

# Morphological, anatomical and molecular analysis of *Coriandreae* tribe (*Apiaceae*) in Turkey

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*Coriandreae* is a small tribe of *Apiaceae* family. In total, two genera and five species are accepted. Four species are native in Anatolia. The genus *Coriandrum* is represented by two species; *Coriandrum sativum* L. and *C. tordylium* (Fenzl) Bornm. The genus *Bifora* is represented by three species; *Bifora americana* (DC.) A.Gray, *B. radians* M. Bieb., *B. testiculata* (L.) Spreng. Misidentification of plants of the genus *Coriandrum* and *Bifora* have found in some herbaria. In this study, easy ways of Coriandreae tribe identification was purposed. All species of tribe, except *B. americana* were collected from different cities of Anatolia. Samples are examined for morphological analysis with a stereo microscope and anatomical analysis with a light microscope. Fruit, stem and leaf cross sections were examined. In addition, molecular analysis studies were also carried out. Plant DNA was isolated from the leaf or fruit. The ITS region is amplified by the polymerase chain reaction. PCR products were sequenced, and phylogenetic trees are generated. Morphological analysis and molecular analysis results were compared. Lack of some parts or unrelated plant identification will result in misidentification. Anatomical information and DNA sequence can be used to confirm the plant species.

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# Environmental niche differences between different ploidy levels in the *Dianthus ciliatus* s. l. (*Caryophyllaceae*)

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Polyploidisation is one of the main evolutionary forces in flowering plants, providing opportunities for their diversification, speciation and adaptation. It is often accompanied by niche evolution of polyploids in relation to their diploid progenitors to avoid interspecific competition in sympatry. Thus, ecological niche shifts including niche expansion or contraction and niche differentiation are among central mechanisms in the establishment of polyploids. We here inferred ploidy levels and their spatial distribution, as well as relative genome size (RGS) variation in Dianthus ciliatus s. l. (Caryophyllaceae) across its native distribution range comprising the Apennine and the Balkan peninsulas. We analysed a total of 84 populations belonging to all three subspecies of the complex, namely D. ciliatus Guss. subsp. ciliatus, D. ciliatus subsp. dalmaticus (Čelak.) Hayek and D. ciliatus subsp. medunensis (Beck & Szyszył.) Trinajsić, of which 67 were included in the RGS analysis. We identified 24 diploid (2x) and 60 tetraploid (4x) populations of *D. ciliatus*, with diploids limited to the Balkan Peninsula. Within diploids, subsp. medunensis had higher RGS values compared to subsp. ciliatus, and within tetraploids subsp. dalmaticus had higher RGS values compared to tetraploid populations of subsp. ciliatus occurring on both the Balkan and the Apennine peninsulas. After identifying spatial segregation in distribution of di- and tetraploids, we compared their environmental niches and tested for niche equivalency and similarity. We hypothesized that polyploids would have a different environmental niche than diploids. In particular, we tested for niche conservatism pattern, the tendency of polyploids to preserve niche traits of their diploid ancestors, which is more expected for autopolyploids than for allopolyploids. Niche analyses showed relatively low niche overlap (26.3%) between the two ploidy levels in the environmental space, with niche of the diploids nested almost completely within the wider niche of the tetraploids. Niche similarity test indicated that the two niches are not significantly conserved, but also not divergent. They differed in their niche components, where tetraploids showed niche expansion (31%) in relation to the diploid niche, with <1% of the diploid niche unfilled. Observed niche expansion of polyploids likely facilitated their dispersal and establishment in novel environments and contributed to their expansion compared to the diploids that remained limited to the Balkan Peninsula.

This work was supported by Croatian Science Foundation project AmphiAdriPlant (UIP-2017-05-2882) and by the Austria–Croatia bilateral project "Genome size evolution and polyploidization in amphi-Adriatic *Aurinia (Brassicaceae)* and the *Cerastium tomentosum* and *Dianthus sylvestris* species groups (*Caryophyllaceae*)" (HR17/2020) funded by the Austrian Agency for International Cooperation and the Croatian Ministry of Science and Education.

#### The use of natural extracts of native plants as an alternative to the use of chemical pesticides in botanical gardens

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The negative effects of chemical pesticides, in the natural environment and in people, are considered one of the main environmental problems on the planet. The negative impact is particularly important in biodiversity, and especially in groups such as invertebrates. As a result of this problem, in recent years, public institutions promoted a policy of banning and restricting the use of chemical pesticides.

As an alternative to these synthetic substances, new products are being developed based on natural substances, which have a much smaller environmental impact.

More than five years the Marimurtra Botanic Garden (Blanes, Girona, Spain) adopted a policy of no use of chemical pesticides to control plant pests. From that moment on, alternatives based on natural products were sought. One of these was the use of aqueous extracts made from plant species with recognized pesticidal or defence-promoting properties. Specialists in this field were consulted and with their advice the garden's technical staff was formed. Also, following his advice, three species were selected to prepare the extracts: *Equisetum telmateia* Ehrh, *Symphytum officinale* L. and *Urtica dioica* L. Recently due to the difficulty in obtaining material supply of *S. officinale*, this has been replaced by *Borago officinalis* L.

Extracts are produced by soaking dry plant material in water. During the maceration time, aeration is periodically applied to prevent rotting. Once the preparation has stabilized, it is strained and stored in cylinders to be used when needed.

From the year 2018, students of the Faculty of Sciences of the University of Girona have the possibility to do their internships or final degree projects in a study to check the effects of plant extracts on cultivated plants. The research consists of growing vegetables in a container, to which plant extracts of each species are applied, plus a mixed one (which includes all three species). During the cultivation time, different parameters are measured: height of the plants, number of leaves, size of the leaves and the presence or absence of pathologies is observed.

In the studies carried out so far it is observed that the effects vary significantly according to the cultivated species but looking at the set of results of all the parameters, it can be concluded that the mixed extract (*E. telmateia* + *S. officinale* + *U. dioica*) is the one that provides the best results.

# Three new species of *Allium* gr. *ampeloprasum* (A. sect. *Allium*, *Amaryl-lidaceae*) from SW-Europe

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The genus *Allium* is one of the most diverse in the world, with more than 1000 accepted species yet (POWO 2023, <u>https://powo.science.kew.org</u>). But a lot of new species are regularly described (several per year), including in the Euro-Mediterranean subgenus *Allium*. Within the section Allium, the A. ampeloprasum polyploid complex is still poorly understood. While a lot of old synonyms are regularly resurrected, new taxa still have to be formally described. He we reinvestigate 3 yet recorded but not formally described taxa, two from S-France (*A. ampeloprasum* sensu Debussche & Debussche; *A. atroviolaceum* sensu Chabert) and one from SW-Spain and Portugal (*A. ampeloprasum* auct. pro parte).

Classical morphology, based both on field and cultivation, and cytology (genome size), are used to characterize these new species from the close related ones, mainly the Atlantic *A. ampeloprasum*, the Mediterranean *A. polyanthum* and the Maghrebian *A. multiflorum*. One of these new species is a rupicolous endemic to Causses (SE-France) and clearly endangered by its extreme rarity. Another is endemic from Atlantic coast of Algarve (from Portugal to Andalucia). The last one is widespread in ruderal habitats and possibly present also in Italy or elsewhere. Its relationships with the also ruderal Eastern Mediterranean "A. ampeloprasum auct." are not well known yet.

### Two new taxa of *Allium* sect. *Pseudoscorodon* (*Amaryllidaceae*) from NE Algeria

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The genus *Allium* is one of the most diverse in the world, with more than 1000 accepted species yet (POWO 2023, <u>https://powo.science.kew.org</u>). But a lot of new species are regularly described (several per year), including in the Euro-Mediterranean subgenus Allium. Within the recently circumscribed section *Pseudoscorodon*, only two species were known in the Flora of Algeria, one of them recently resurrected. During field trips on poorly known areas of Bibans and Djebel Maadid we encounter unusual plants belonging to the genus *Allium*.

The first one is very close to the Algerian narrow endemic *A. trichocnemis*. In spite of the enlarged distribution area than previously given for the latter, our plants still grow outside the range and show slight differences on the flowers. Thanks to the vegetative characters, strongly identical to those of *A. trichocnemis* s. s., and diverging with the close related *A. seirotrichum* (another narrow endemic), we propose to described this new taxon as a subspecies of *A. trichocnemis*.

The second one is very distinct of any Algerian Allium. It belongs to the *A. rouyi*'s group, known from Spain (3 species), Morocco (*A. valdecallosum*) and Tunisia (the recently discovered *A. elaounii*). All these species are poorly known, with few or none colour photographs and poor data about the leaves at green stage. Nevertheless, our data, both from field and cultivation, suggested us that our plants belong to a new taxon. Because the phylogenetic relationships between all these taxa are not known, we chose to describe it at species rank, waiting for more information.

Both the new species and the new subspecies are very scarce, presumably narrow endemic and at least one of them clearly endangered. Their consideration will now reinforce or confirm the classification of both Bibans and Djebel Maadid as Important Plant Algeria (Key Biodiversity Areas for plants).

# Anatomical and morphological investigation on endemic *Alkanna cappadocia* (*Boraginaceae*)

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*Alkanna*, a plant native to the Mediterranean, has about 40 species in the world. There are 31 species in Turkey, including *Alkanna cappadocica* Boiss. & Balansa., which is one of the endemic species. *Alkanna* genus is important in Turkey in terms of endemic species. *A. cappadocia* is a greenish perennial plant that grows on dry slopes and steppes at altitudes of about 1100–1700 m in the provinces of Nevşehir, Kayseri and Adana, and its height varies between 15–40 cm. It was dried at room temperature in an opaque drying chamber. In addition, some samples were taken into 70% ethanol for the anatomical and morphological examination of the collected plant parts. The morphological and anatomical research on *A. cappadocica* is described in the current study.

While anatomic-morphological thinning was done, transverse and superficial sections were taken from both basal leaves and leaves, and transverse sections were taken from the stem and root. At stem layer of cuticle is thin. Epidermal cells are circular, have thin walls, and only one layer. The leaves and stems have mostly short glandular hairs and common set of setiform eglandular hairs ranging in size from 1–3 mm. The eglandular hairs of the base leaves are longer and the cuticle is dotted. Leaf is monofacial. On both the top and bottom surfaces of leaves, a thick cuticular layer is present. A single line of cells with about equal breadth and length makes up the top and lower epidermis. Thin cuticle layer. Epidermal cells of leaf is oval, single-layered, and having thin walls. The root is perennial. On the other side of the root, the periderm is multilayered. Phellem's components are brown. There are 15–20 layers in the parenchymatic cortex.

### Seed orchards and clone collections as a tool for *ex situ* conservation of the genepool of Bulgarian conifers

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The effective conservation and use of forest genetic resources requires a combined approach consisting of both in situ and ex situ activities. The in situ measures are straightforward - they are focused to protection (in broadest sense) of natural populations, including applying of a special management regime. Ex situ approach is more sophisticated, more laborious and more expensive. However, it is irreplaceable in the cases of destroyed natural environment of the target populations. The approach consists of several groups of measures ranging from establishment of artificial populations and clone collections to storage (including cryo-storage) of seeds, pollen and other propagules. Forest seed orchards and clone collections provide an additional opportunity for ex situ conservation because they are part of breeding activities and their establishment and maintenance can be economically feasible. Capturing of the existing genetic diversity in the natural populations for ex situ conservation is of crucial importance for the success of the conservation efforts and could contribute to optimizing between gain and diversity - the never-ending dilemma of the tree breeder. Considering the effective population size concept (Ne) could help the representative sampling for ex situ conservation. This concept should be applied considering the life-history characteristics of the target species, and particularly, the mating system and natural propagation.

Conifers represent a group, important both from economic and conservation viewpoints. There are about 15 coniferous species occurring naturally in Bulgaria and most of them are considered important due to various reasons. More than 40 seed orchards and about 20 clone collections have been established during the last 50 years for species such as *Pinus sylvestris* L., *P. nigra* Arn., *P. peuce* Griseb., *Picea abies* (L.) Karst., *Abies alba* Mill., but also for some species considered to be of importance as medicinal and aromatic plants, like *Juniperus communis* L. and *J. oxycedrus* L. s. l. Most of these seed orchards and clone collections represent valuable genepool. However, they need urgent measures for inventory and proper management for improving their effectiveness as ex situ conservation objects.

### *Centaurea dolopica (Cardueae, Asteraceae)*, a new species of section *Acrocentron* from Greece

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A new species of Centaurea section Acrocentron (Asteraceae) from Greece is described and illustrated based on morphological and molecular evidence. Centaurea dolopica sp. nov. was collected from Omvriaki area in Central Greece whereas the specific epithet refers to the ancient name of the region (Dolopia). It belongs to the Greek endemic group including Centaurea achaia, C. aetolica, C. corinthiaca and C. euboica (C. achaia group), but differs morphologically from the other members mainly by its narrower involucral bracts, its fewer central florets and robust habit with larger, more branched flowering stems bearing a larger number of capitula. The species is restricted to a single locality where its small population grows along roadsides and rocky cut slopes. Individuals of Centaurea dolopica were also examined karyologically resulting to the chromosome number 2n = 2x = 22 accompanied by comprehensive karyotype analysis. Phylogenetic study including the new species and major representatives of Centaurea section Acrocentron was conducted using two nuclear markers (ITS, ETS). The results revealed the placement of Centaurea dolopica in the Greek-Aegean clade of the section in sister relationship to the representative of C. achaia group, corroborating the morphological and karyological data. Based on the current data and IUCN criteria, the species should be listed as Endangered (EN).

#### The use of chemical composition of the essential oil as taxonomic markers – a case study with five species of the genus *Cyanus (Asteraceae)*

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*Cyanus* Mill. (*Asteraceae*) is a well-defined and at the same time taxonomically complicated genus. There are still different opinions about its status - an independent genus or a section of the genus *Centaurea* L. According to The Plant List (2023), *Cyanus* includes 50 accepted taxa distributed throughout Central and Southern Europe, North Africa, Asia Minor, and the Caucasus, with some species appearing as far east as Iran and Afghanistan. The majority of species are perennial, divided into 2 groups: *C. triumfettii* gr. with cord-shaped, unthickened roots and *C. napulifer* gr. with rump-like or tuber-like thickened ones. Less than 0.5% of the species are annuals or biennials. Despite increased interest and focused research in recent years, there is still much ambiguity regarding the boundaries between outlying taxa and their rank.

To solve these problems, researchers simultaneously use different approaches and methods (morphological, karyological, genome size, molecular, etc.). The present study aims to show that the information on the chemical composition of the essential oil also has a great potential to be used as an additional method for the needs of taxonomy. For this purpose, a comparative analysis of the chemical composition of the essential oil of five species of the genus, evaluated by GC-MS, was made: 3 perennial – *C. adscendens* (Bartl.), *C. orbelicus* (Velen.) Sojak (from Bulgaria), *C. tuberosus* from Croatia and 2 annual species – *C. segetum* Hill and *C. tchihatcheffii* (Fisch. & C.A.Mey.) Wagenitz & Greuter from Turkey.

In the metabolic profile of the representatives of *C. napulifer* gr. oxygenated sesquiterpenes predominate, accounting for almost 50% of all compounds identified. In the perennial representative of *C. triumfettii* gr. sesquiterpene hydrocarbons predominate (over 50%), in annuals, oxygenated monoterpenes are best represented, and in *C. tchihatcheffii* a significant amount of oxygenated sesquiterpenes is also observed.

The obtained results confirm the hypothesis of the successful use of certain compounds as chemotaxonomic markers in the genus *Cyanus*.

This work has been carried out in the framework of the project "Modern trends in the flora, vegetation and natural habitats of Bulgaria and the Balkan Peninsula" of the Institute of Biodiversity and Ecosystem Research, Bulgarian Academy of Sciences.

# The impact of climate change on the phenology of selected species of the Bulgarian flora

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There is scientifically based data that climate is warming in the past century. In plants, climate change inevitably affects their phenology, leading to a shift in the phenological phases. Hundreds of thousands of specimens collected in the last 100 years are kept in the collections of the Bulgarian herbaria - SO, SOA and SOM. Their use as "live images" of phenological events is particularly suitable for characterizing the phenological responses of plants under the influence of climate. The aim of the present study is to trace the flowering time of selected species of the Bulgarian flora during the last century in order to answer the question of whether and how climatic change on the territory of Bulgaria has affected their phenology. Three species of conservation concern with single or a few localities in the country were selected - Astracantha arnacantha subsp. aitosensis (Ivan.) Reer & Podlech, Calystegia soldanella (L.) Roem. & Schult., and Rhododendron ponticum L. All species are distributed in the Black Sea biogeographical region, which covers just 0.3% of the EU territory. All available specimens of the target taxa were studied in detail, and data were taken on the specific localities, the date of collection and the phenological phase of the collected plant individuals. The arithmetic mean of days with full bloom was calculated for data before and after 1950. Data were processed with specialized statistical software Sigma Plot. The results of the study show that in the three target species there is a shift of the flowering period forward by several days, i.e. earlier flowering, with Calystegia soldanella being the most significant example. Additionally, it can be noted that by conducting similar type of observations and analysis, valuable data can be provided on the pace and direction of the impact of climate change on plant species.

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#### Index

Aberkane H83
Aceituno-Mata L72
Acquaviva R
Adamčík S110
Addink W67
Agrawal S.K
Ahmed Amri A
Akalın E
Al Tawaha A.R.M.S
Aleffi M
Álvarez I
Anadei L
Aneva I
Apostolopoulos E
Aprile S
Aquilanti L40
Arroyo J48
Asens G
Astuti G66
Bacchetta G19, 37, 39
Badalamenti N133
Bajona E21, 101, 117
Baldemir Kılıç A130
Ballar Kırmızıbayrak P79
D 1 G 100 100
Bancheva S
Bancheva S
Barazani O85
Barazani O85 Barblishvili T102, 115
Barazani O
Barazani O.
Barazani O
Barazani O
Barazani O.
Barazani O.       .85         Barblishvili T.       .102, 115         Barone G.       .60         Bartoli Fi.       .117         Bartoli Fl.       .22         Bartolucci F.       .37         Barucca2 M.       .40         Başer K.H.C.       .15, 75         Bassi F.       .83         Baumel A.       .16, 37         Bazos I.       .103         Bedir E.       .79         Bellanca L.       .118
Barazani O.
Barazani O.       .85         Barblishvili T.       .102, 115         Barone G.       .60         Bartoli Fi.       .117         Bartoli Fl.       .22         Bartolucci F.       .37         Barucca2 M.       .40         Başer K.H.C.       .15, 75         Bassi F.       .83         Baumel A.       .16, 37         Bazos I.       .103         Bedir E.       .79         Bellanca L.       .118         Benítez G.       .72         Berežni S.       .77
Barazani O.
Barazani O.
Barazani O.
Barazani O.
Barazani O.

Bonanno F	
Bosch-Guiu A.	92
Boschin M	
Bossdorf O	65
Bou Dagher Kharrat M	16
Breidy J.	
Brundu G	89
Bruno M.	
Bucchini A.E.A.	80, 81
Burguera i Martín C.	104
Caboň M.	
Caboňová M.	110
Caković D.	
Camarda I	23
Campisi P	29
Cantonati M.	.43, 44, 45
Caputo P.	
Caralt F	92
Carballal R	
Carducci F	40
Carine M.	
Carlos Gómez-Bellver C	88, 92
Carrion G.	92
Caruso T	113
Casavecchia S.	40
Casazza G	
Cetlová V.	110
Chandler K.	68
Chatelain C.	64
Chatellier C.	54
Cianfaglione K.	
Cid Rodriguez M.	44
Cogoni A.	
Constantinidis T99	, 100, 107
Conti E	49
Conti F	37, 39
Coppi A	101
Cornara L.	76
Cristofolini G	61
Culshaw V.	
Dafni A	71
Dagnino D	17
De Agostini A.	24
De Feo V.	76
De Luca D	

del Tio Navas C.	57
Deussing L.M.	112
Di Giacomo C.	78
Di Gristina E21,	60, 119
Di Guardo M.	16
Diadema K.	16
Dimopoulos P.	132
Dokmak H.	
Dolci D.	
Domina G21, 39, 60, 1	14, 119
Dörflinger G.	44
Dorfner M.	
El Mokni R	34
El-Bouhssini M.	83
Epifanio C.S.	112
Fàbregas J.	127
Fačkovcová Z.	
Falace A.	
Farris E	19, 53
Feliu P	
Fernández-Mazuecos M.	
Fior S	
Fitz Patrick U.	68
Fois M.	19
Fraga i Arguimbau P55, 93, 104, 1	123, 127
Frajman B.	.34, 126
Franzoni J	
Fuertes-Aguilar J.	
Gabedava L.	
Gajdošová Z.	
Galgóci M.	
Garcia S32,	
García-Fernández A.	
Gargano M.L.	94
Garnatje T.	32
Garnatje T Garriga S	
	123
Garriga S.	123 .50, 110
Garriga S Gbúrová Štubňová E Giacò A Giacobbe S	123 .50, 110 37, 52 116
Garriga S. Gbúrová Štubňová E. Giacò A. Giacobbe S. Giamperi L.	
Garriga S. Gbúrová Štubňová E. Giacò A. Giacobbe S. Giamperi L. Gianguzzi V.	
Garriga S. Gbúrová Štubňová E. Giacò A. Giacobbe S. Giamperi L. Gianguzzi V. Giannelli F.	
Garriga S. Gbúrová Štubňová E. Giacò A. Giacobbe S. Giamperi L. Gianguzzi V. Giannelli F. Giarratana S.	123 .50, 110 37, 52 116 80, 81 60 40
Garriga S. Gbúrová Štubňová E. Giacò A. Giacobbe S. Giamperi L. Gianguzzi V. Giannelli F. Giarratana S. Gibernau M.	
Garriga S. Gbúrová Štubňová E. Giacò A. Giacobbe S. Giamperi L. Gianguzzi V. Giannelli F. Giarratana S. Gibernau M. Giovannotti M.	
Garriga S. Gbúrová Štubňová E. Giacò A. Giacobbe S. Giamperi L. Gianguzzi V. Giannelli F. Giarratana S.	

Grdzelishvili I115
Guarino C59
Guerrina M17
Guirado M
Gutiérrez-Larruscain D
Guttová A
Harpke D
Hidalgo O32
Hodgetts N26
Ibáñez N
Iocchi M22
Iriondo J.M56, 57
Isocrono D120
Ivan Rúrik I50
Kantor A
Karacabey E
Karacaocy E
Khadari B
Kilian N
Klobučník M110
Kochjarová J50
Koşar M75
Koskina C
Kotsabas K100
Koutroumpa K
Kozuharova E
Kučera J
Küçüksolak M
La Malfa S
Lachashvili N
Lambevska-Hristova A134
Lange-Bertalot H44
Lara-Romero C56, 57
Lazzaro L101
Lechner H
Liveri E
Ljubičić I20, 126
López-Guillen E
López-Pujol J
Lozano V
Lucchese F
Maccioni A
Maccioni S
Macis S53
Magdy M122
Magrini N66
Maguilla E48
-

Malacrinò V.	
Malaspina P	
Malfa G.A.	
Mandáková T	
Manghisi A.	
Mannino A.M.	91
Manoylov K.M.	43
Manzo E	
Marchese A.	
Marcial Escudero M	
Marhold K.	
Maria Guerrina M	
Marino P78	
Marra F. P.	
Mazzola P	
Médail F.	· · · · · · · · · · · · · · · · · · ·
Medina N.G.	
Melero R.	
Melia N.	
Melichárková A.	
Miceli N.	
Mimica-Dukić N	
Minuto L.	
Mirabile G	
Molina A.	
Molina J.	
Mombrial F.	
Mondello F.	
Morabito M.	
Morcillo J.	
Mucko M.	
Mugnai M.	
Muñoz J.	
Muñoz-Rodríguez P	
Munzi S.	
Nakhutsrishvili G.	
Niek Scheepens J.F.	
Nieto Feliner G	
Nieto Feliner G.N.	
Nualart N Oberprieler C	.00, 92, 108, 111
Obré J.	
Olsson S	
Orčić D.	
Orsenigo S.	
Ouahmane L	16

Pagès J.M.	92
Pahl J	112
Palla F	
Paoli L	
Papatheodoulou A	44
Pardi R	94
Pardo-de-Santayana M	
Pascual-Díaz J.P	
Paun O	50
Pausas J.G	
Pellicer J	32
Peruzzi L33	3, 37, 39, 52, 66
Pesaresi S	40
Pickering V.	105
Pierotti F	66
Podda L	
Pokorny L	18
Polito F	76
Poponessi S	
Prigioniero A	
Puglisi M	
Quattrini G	
Raimondo F.M. 21, 76, 8	36, 98, 114, 117,
118, 119, 121	
Raimondo G	
Ranauda M.A	
Rat M	
Ravera S	
Rebbas K	
Rejdali M	
Rešetnik I	
Riina R	
Rincón-Barrado M	
Rindi F	
Rizk S.M.	
Rodríguez-González R	
Rodríguez-Prieto C	
Roma-Marzio F	
Ros R.M.	
Rosato M.	
Rosselló J.A.	
Rubio Teso M.L	
Saber A.A Sacristán-Bajo S	
Sacristan-Bajo S Sáez L	
Saez L Salerno F	
Salerno F Sanchez-Garcia M	

Sanguin H.	16
Sanmartín I.	
Sans A.	124
Scarano G.	
Segarra-Moragues J.G.	25
Selvi F.	101
Senko D.	
Simón-Porcar V.	48
Šingliarová B	
Skokanová K.	
Skubic M.	34
Šlenker M.	
Slovák M.	.28, 50, 110
Smyth N.	
Sönmez S.	
Sottile S.	
Spadaro V.	
Spagnuolo D	
Španiel s	
Spitale D.	
Stancheva S.	
Stefanaki A.	
Stout J.	
Svitok M.	
Tartaglia M.	
Tasheva-Terzieva E.	
Tchanturia-Kvirtia T.	
Temunović M.	
Terlević A.	
Tiburtini M.	
Tison J.M.	
Tollon C.	
Tomasello B.	
Tomasello S.	
Torices R.	
Torres E.	
Torroella V.	
Trucchi M.	
Üner G.	
Valdés B.	
Valdés-Florido A.	
Valenzuela L.	
Vallès J.	
van Andel T.	
Vangelisti R.	
Varaldo L.	
Vargas P.	
, u1 5 u0 1	

Vdačný P	50
Véla E	
Véla Ouzzani S	128
Venturella G.	119
Viciani D.	101
Views C.	
Villaverde T.	
Viruel J.	16
Vitales D.	32, 92, 111
Vladimorov V.	134
Werner O	122
Willems F.	65
Yazbek M.	
Yılmaz S	79
Yusufbeyoglu S	130
Zangari G	22
Záveská E	
Zhelev P.	
Zikos A	103
Zizzo G	
Zografidis A	132
Zozomová-Lihová J	31
Zumel D	122
Zuzolo D	59









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