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ABSTRACT BOOK

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Biodiversity, ecosystem conservation and climate resilience: bridging science and action

KEYNOTE

Conservation paleoecology: how the past can help the future

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Our planet is currently experiencing dramatic changes in climate, land cover, biodiversity abundance and distribution in response to anthropogenic land-use, resource consumption, industrialization, and intensive production. Action is needed to counter climate disruption, extinctions, loss of diverse ecosystems, pollution. Otherwise, driving Earth's life support systems towards a dangerous tipping point. Studying how the ecosystems functioned in the past, before human activities were widespread on Earth, will allow us to establish guidelines for conservation strategies based on investigations of past ecosystems. This talk will address three key conservation issues where knowledge from the past can help the future: (1) helping saving endangered species by investigating their fundamental niche, (2) helping restoring disfunctioning ecosystems by identifying missing keystone megafauna species and reinstating them or adequate surrogates, and (3) helping to be better prepared for a warmer future by looking at ecosystems during warmhouse periods in the geological past.

Keywords: conservation; fundamental niche; keystone species, surrogate species; climate warming.

KEYNOTE

Pathways to coexistence with large carnivores in Romania

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Our society is dependent on persistent economic growth and often dramatically transforms the environment. Romania's large carnivores (European brown bear, grey wolf, Eurasian lynx) have large abundances when compared with other European countries and a complex spatial ecology, with large home range sizes (e.g., over 3000 km2 for bears) overlapping human-modified landscapes, which increase interactions that escalate into conflicts. These conflicts are often exacerbated by decreasing tolerance to large carnivores and new policies favouring the development of wildlife-rich areas. For the brown bear, the species that generates the most conflicts in Romania, the current management system primarily is based on on lethal control of the population (trophy hunting, hunting of nuisance bears), ignoring other measures such as bear-proof waste management to prevent habituation, prevention of degradation and fragmentation of habitats, bear-friendly tourism, and management of livestock. Moreover, the reliance on lethal control has been largely arbitrary, as there has been no assessment of its effectiveness in reducing conflicts. Grey wolf is another conflictual species; however, it is much easier, as only livestock management is usually necessary. Still, lethal control of the population is favoured and will soon become a norm, as its status was downgraded at the European level from strictly protected species to protected species. To safeguard both people and wildlife, environmental authorities should adopt the concept of mutual adaptations between humans and wildlife in shared landscapes and develop holistic responses co-implemented with the most affected local communities, and supported by systematically collected and credible evidence.

Keywords: european brown bear; grey wolf; human-wildlife conflicts; wildlife management

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AVISEN: AVIan SENescence database to understand senescence in birds

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Senescence is the biological process of progressive damage accumulation and physiological dysfunction, leading to a gradual deterioration of organismal health and a decrease of survival prospects. Studies focusing on model organisms have provided molecular insights into the processes of senescence within species, while analyses on broader taxonomic scales using wildliving organisms have shown that senescence patterns can be highly variable. Of special interest in this context are birds, because of their long lifespan and slower senescence compared with size-matched mammals, despite several physiological and cellular avian traits (e.g., high blood glucose levels, metabolic rates and body temperatures), which were expected to contribute to senescence. This phenomenon is known as the 'bird paradox'. Making use of increasing empirical data on senescence in wild birds, we compiled the AVIan SENescence (AVISEN) database. This extensive literature review yielded longitudinal and cross-sectional data about age-specific reproduction, survival and phenotype. AVISEN currently contains 923 speciesspecific datasets of 101 bird species and 142 populations, derived from 199 scientific publications covering most of the global biogeographic regions. AVISEN includes 659 reproductive, 73 survival and 191 phenotypic life tables. Preliminary analyses indicate evidence of senescence in 54% of reproductive and 46% of phenotypic datasets. We present preliminary results on the influence of ecological traits on senescence in birds.

Keywords: age-specific life tables; bird senescence; comparative aging studies; wild animals

The fine structure of the antennae in *Capnodis cariosa* (Coleoptera: Buprestidae) and their role in populational distribution.

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Capnodis cariosa, the largest buprestid beetle in Europe, was found in the Southern Dobrogea (SE Romania) until 1967, the last known record. A new record was in 2015, but in the SW Romania, in the Danube Defilee. We found the presence of a consistent population in this area, and we monitored this population during 2023 and 2024. Additionally, we observed the presence of beetles in the same bushes as its hostplant, *Cotinus coggygria*, during those investigations. Therefore, we studied the antennae of a male and female to investigate the ultramicrostructure and the types of sensilla and their positions on the antennae and then speculated how the antennae work in the individuals' grouping.

Diversity and distribution of weevils (Coleoptera, Curculionoidea) in the Botanical Garden "Alexandru Borza" from Cluj-Napoca

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The Botanical Garden "Alexandru Borza" is located close to the central area of Cluj-Napoca, one of Romania's largest urban areas, and therefore the beetle fauna could be affected by anthropic influence. The area was divided into five sectors, roughly following the main sectors of the garden. Weevils were collected from those five regions, between April-October 2024. We identified individuals from 38 species, 27 genera, 6 subfamilies, and 3 families. Before our research only two species were found, Gymnetron rostellum Herbst, 1795 and Mononychus *punctumalbum* Herbst, 1784 (Marcu, 1957). Only the latter was present in the collected material. The other 37 species were spotted for the first time in the Botanical Garden. Surprisingly, the species from Apioninae subfamily were predominant, even though the general trend for it is to be placed after the Entiminae subfamily. This unexpected species distribution could be caused by the preference of many species from the Apioninae subfamily to consume herbaceous plants, a factor that could have shielded them from potential threats. The highest weevil diversity was recorded in the second sector (32 species), followed by the third (20 species), first (10 species), fourth (7 species), and fifth (5 species) sectors. The dominant species were Zacladus exiguus Olivier, 1807 (31.32%), on Geranium sp., and Protapion apricans Herbst, 1797 (15.85%), on Trifolium pratense. Remarkable for the Botanical Garden was the presence of the: Liophloeus (Liophloeodes) lentus Germar, 1823 and L. (s.str.) tessulatus O. F. Müller, 1776, found on Aegopodium podagraria. We found 3 rare species: Exomias chevrolati Boheman, 1843 and Sibinia phalerata Gyllenhal, 1835 in the second sector, and Smicronyx coecus Reich, 1797 in the first sector. We did not find the expected species richness of weevils. Probably those beetles are affected by insecticides that are used to keep the insect communities under control and preserve the plants.

Keywords: weevils, faunistic studies, host plants, rare species, ecology

Testing new AI-based automated methods to monitor pollinators in "Eastern Cluj Hills" area (Cluj, Romania).

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The rapid decline of pollinators populations has led to calls for developing improved, costeffective and automated as possible methods for simultaneously monitoring plants, pollinators, and their interactions across space and time. A standardized monitoring throughout Europe would allow for trends in pollinators and in the ecosystem service of pollination to be detected in near time, allowing for rapid management interventions. New tools for detecting, identifying, and counting insects using machine learning and computer vision are in the later stages of development. The project "Standardized European monitoring of plant-pollinator interactions" (SEPPI) proposes to advance this research beyond the state-of-the-art by developing and revising protocols using AI tools for automated detection and identification of floral-visiting insects. In 2024, SEPPI team developed a protocol that was tested in eight European countries: Germany, Belgium, Hungary, Czechia, Latvia, Finland, Italy, and Romania. The protocol involves assessment of differences in pollinator diversity and composition and in plant-pollinator networks between traditional versus automated methods across spatial environmental gradients. For the automated sampling we used non-lethal technology to capture images of pollinators on flowers in the field using time-lapse cameras. In Romania, we implemented the protocol and evaluated both automated and traditional methods in the Eastern Hills of Clui area. The assessment was conducted across nine sites representing various land management types: abandoned meadows, hay meadows, and moderately sheep-grazed areas. The preliminary results suggest that the automated pollinator monitoring method is more time efficient compared with traditional method. This approach is easy to apply in the field and significantly reduces human workload. Once the new AI tools will be trained to identify the pollinators, it could provide standardized methods that can be scaled up without requiring previous taxonomic knowledge. Also, the new AI tools have the potential to be extended to various ecological studies.

Keywords: Artificial Intelligence, automated monitoring, plant-pollinators interactions.

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Gall formation on wild roses under laboratory conditions: roles of wasp origin and host susceptibility"

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Rose gall wasps (Diplolepididae) are specialized insects that induce gall formation on young shoots of wild roses (*Rosa* spp.). Our laboratory experiments aimed to investigate the process of gall formation under controlled conditions. Using the species *Diplolepis rosae* and *D. mayri*, we examined the dynamics of gall growth, the role of oviposition frequency, and the susceptibility of different rose species. Oviposition events were observed within a daily 1-hour window, and the number, size, and location of galls were documented. Our results showed that the population origin of the gall wasps influences the speed and size of gall development. Higher oviposition frequency led to a significantly greater number of galls, predominantly forming on the plant stems. Furthermore, different wild rose species exhibited varying degrees of susceptibility: some species were more resistant, producing fewer and smaller galls, while others were more susceptible, forming larger and more numerous galls. This suggests that the physiological characteristics of host plants affect gall formation. In conclusion, we found that gall development is regulated by complex interactions involving the origin of the wasps, the intensity of oviposition activity, and species-specific traits of the host plants. Our findings contribute to a deeper understanding of plant-insect interactions and the potential industrial and medicinal applications of galls.

Keywords: *Diplolepis rosae*; *Diplolepis mayri*; oviposition frequency; plant-insect interactions; gall ecology.

Acknowledgements: We thank Balázs Robert Zoltán for his help with the rose growth facilities and Mátis Attila for the identification of wild rose species.

Towards an integrated understanding of vegetation dynamics in the Carpathians

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High-elevation ecosystems above the treeline in the Carpathian Mountains have been shaped by seasonal pastoralism since the Bronze Age. In recent decades, these landscapes have undergone significant changes, largely driven by socio-economic shifts. Regional-scale insights into vegetation dynamics, integrating ground and remote sensing data, remain limited. To address this gap, we conducted research as part of several collaborative projects. Our approach integrates advanced analysis of long-term satellite imagery, multi-year microclimatic monitoring, diachronic analyses of historical aerial photographs, and dendrochronological investigations to reveal shrubland shifts beyond satellite records. Together, our findings have documented a significant increase in vegetation greenness over the last four decades, mainly driven by the expansion of mountain shrubs in formerly grazed lands. Our ongoing investigations aim to examine the climatic and socio-economic drivers of these changes and their consequences for biodiversity. This integrative framework aims to provide critical insights into the ecological trajectories of high-elevation Carpathian ecosystems under ongoing environmental change.

Keywords: Carpathians, greening, remote sensing, microclimate, dendrochronology.

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Integrating allometry and digital image analysis for *Prunus tenella* Batsch. leaf phenotyping

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Accurate and efficient measurement of leaf traits is crucial for advancing plant phenotyping and ecological research. Prunus tenella Batsch, a crop wild relative of the cultivated almond, offers unique opportunities for such studies. Phenotyping and characterizing adaptations across populations in their native environments are vital for conservation, assessing climate change adaptability, identifying local adaptations, and supporting crop improvement efforts. However, traditional methods for measuring traits are often time-consuming and labor-intensive. We developed an integrated phenotyping workflow combining allometric modeling and digital image analysis (DIA) to rapidly estimate leaf area and leaf dry weight, key traits of the leaf economic spectrum. We analyzed 958 leaf samples from 11 populations spanning Romania, Hungary, and the Kosovo region. Three digital image segmentation approaches were evaluated and compared: (1) manual ROI-based leaf area measurement on each leaf in Image] (reference method), (2) automated fixed threshold segmentation in Image on the image set (3) automated algorithm-driven foreground/background segmentation with AI Photoroom-Pro. For each leaf, we recorded direct measurements of area and dry weight, and developed allometric models to predict dry weight from image-derived leaf area. Our results show strong correlations (R²>0.84) between predicted and measured dry weights, and high agreement across DIA methods, with automated, fixed thresholding (RMSE=3.08) performing better than Photoroom-AI (RMSE=35.78) in measuring leaf area as our reference method, thus both being appropriate to estimate leaf dry weight via linear modeling. This combined digital and modeling strategy enables rapid, high-throughput, and reliable estimation of leaf biomass, providing a feasible alternative to laborious direct measurements and supporting large-scale, field-based studies of adaptation. To our knowledge, this is the first study to integrate digital image analysis with species-specific allometric modeling to predict leaf dry weight from image-derived measurements in *Prunus tenella*, enabling a novel, scalable methodology for rapid phenotyping of functional traits across its range.

Keywords: allometric modeling; crop wild relatives; digital image analysis; ImageJ; phenotyping

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Seed bank as a source for the spontaneous regeneration of dry grasslands on former arable fields in an agro-pastoral landscape

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Natural and semi-natural dry grasslands in Europe have suffered significant losses in both extent and species richness due to overgrazing and conversion into croplands. Abandoned crop fields and field margins offer the possibility for grassland restoration, especially if environmental policies encourage habitat rehabilitation. This study investigates seed bank diversity and density across a succession gradient on abandoned crop fields, unravelling the role of seed banks in spontaneous grassland regeneration. In addition, as a more practical aim, we developed biological trait-based profiles of grassland specialists with transient seed banks. We assessed seed bank composition and above-ground vegetation in young, medium aged and old abandoned arable fields respectively old-growth grasslands at three sites in western Transylvania (Romania). Our analysis revealed that while seed bank richness was much lower than above-ground vegetation at the sample level, at the landscape level, half of the species were represented in the seed bank as well. In addition, the seed bank of both abandoned fields and old-growth grasslands was dominated by grasslands specialists. Furthermore, we found a unimodal relationship between the species richness of the seed bank and successional stage. This is potentially attributed at the ascending part of the curve to the seed rain of establishing species at younger stages, while at the descending part to seed viability loss, declining abundance of ruderal species, and litter accumulation over time, which prevents seed penetration into the soil. Furthermore, based on our trait analysis, slow colonising grassland specialists with transient seed banks typically exhibit a lack of any specialized dispersal appendages and have insufficient clonal dispersal mechanisms. Our results support relatively high grassland regeneration potential from the seed bank in marginal abandoned fields at the landscape level, while suggesting, that slow colonizing grassland species with transient seed banks should represent the main targets for reintroduction.

Keywords: abandoned fields; classification tree; grassland specialists; restoration; trait analysis

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Visitors' perceptions of Bucovina on Instagram: which places generate more social media engagement?

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Instagram is one of the most influential social media platforms, shaping the way tourists imagine and experience destinations. It is currently used as an instrument for influencing visitor behavior and to increase tourist flows. This paper analyzes the spatial distribution and social media engagement of Instagram posts tagged with the label #discoverbucovina. A number 6.472 Instagram posts were used to identify the most popular places associated with Bucovina on Instagram, and especially the weight of rural and urban places and heritage in people's posts. Even if Bucovina is well known as a traditional rural destination, the results indicate a strong concentration of the Instagram posts in the most important cities and in several popular rural destinations that gather the main tourist attractions and facilities in the region. The distribution of posts is also linked to accessibility, tourism tradition and specialization. An east-west gradient was observed in the distribution and engagement of Instagram posts, also linked to the relief altitude. As people's posts reflect their perceived importance of places and heritage, further influencing other visitors, this study is particularly relevant for a better capitalization and management of resources in this region, as well as for other rural and nature (mountain)-based destinations.

Keywords: social media; tourism; rural areas.

Molecular biology for sustainable health

KEYNOTE

Genetic control of chloroplast biogenesis: focus on the plastid RNA Polymerase complex and PAPs

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The chloroplast is the semi-autonomous organelle of eukaryotes that performs photosynthesis. In higher plants, chloroplast biogenesis depends on a tight transcriptional coordination of both nuclear-and-plastid photosynthesis-associated genes. The plastid-encoded RNA-polymerase (PEP) is composed of a plastid-encoded catalytic core, similar to multi-subunit RNA polymerases, bound to fifteen nuclear-encoded PEP-associated proteins (PAP1- PAP15). The binding of all the PAPs to the catalytic core is essential for chloroplast biogenesis. The structure of the PEP from Mustard will be presented along with the proteins that are in the vicinity of PAP8s in Arabidopsis. Spatial organisation, protein trafficking and light signalling will be discussed.

Keywords: Chloroplast – PAPs – PEP - Photoreceptors.

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Novel Tumor-Targeted Nanoformulations Containing Simvastatin and Doxorubicin Suppress Melanoma Progression in a Murine Model

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Melanoma is one of the most aggressive cancers worldwide, rapidly developing resistance to conventional therapies, which often results in metastasis and recurrence. This study investigates a novel therapeutic approach targeting both tumor-associated macrophages (TAMs) and cancer cells using IL-13-PEG-LCL-SIM and PEG-EVs-DOX. These nanoformulations were evaluated for their potential to reduce the aggressiveness of B16.F10 murine melanoma. Mice bearing melanoma tumors received intravenous administration of either the combination therapy or individual formulations of SIM or DOX. Angiogenic protein expression was analyzed using a protein array, while levels of apoptotic markers Bcl-xL and Bax were assessed via Western blot. High-performance liquid chromatography (HPLC) was used to quantify malondialdehyde (MDA), a biomarker of oxidative damage, in tumor lysates, Additionally, matrix metalloproteinase-2 (MMP-2) activity was assessed as an indicator of tumor invasion and metastasis. The combination treatment group exhibited a strong suppression of tumor growth (94%). Compared to controls, this group also showed decreased expression of several proangiogenic proteins under the targeted therapy condition. Notably, MDA levels were significantly elevated in the combination treatment group, suggesting enhanced disruption of intratumoral reactive oxygen species (ROS) homeostasis. Despite the observed therapeutic effects, none of the nanoformulations reduced MMP-2 activity. However, the combination treatment led to an approximate 1.5-fold increase in the Bax/Bcl-xL ratio, indicating the induction of a pro-apoptotic state. These findings support the conclusion that the actively targeted combination therapy effectively suppresses tumor progression. This novel drug delivery strategy based on combined active targeting of both cancer cells and immune cells was able to induce a potent antitumor effect by disruption of the reciprocal interactions between TAMs and melanoma cells.

Keywords: melanoma, targeted therapy, tumor microenvironment, extracellular vesicles, liposomes

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Development and characterization of curcumin-loaded extracellular vesicles as an adjuvant to anti-PD-L1 therapy on melanoma 3D model

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Malignant Melanoma is currently one of the most aggressive types of cancer due to its invasive and metastatic potential. Conventional therapies such as radiotherapy and chemotherapy have not proved to be efficient, having systemic side effects. Newly developed immune checkpoint inhibitor therapies show promising results, but the resistant phenotype of melanoma still renders them ineffective in some cases, underlying the necessity for novel approaches. This study focuses on developing an immunostimulatory therapy based on curcumin-loaded extracellular vesicles (EVs-CURC) derived from Trp-2/CpG-ODN (Tyrosinase Related protein-2/Cytidine monophosphate guanosine oligodeoxynucleotides) pulsed dendritic cells, in combination with an antibody that functions as a PD-L1 immune checkpoint inhibitor, to enhance the immune response against melanoma cells. We evaluated the efficacy of this therapy using a heterocellular spheroid that recapitulates the immunosuppressed melanoma microenvironment. Extracellular vesicles derived from dendritic cells (DC2.4) activated with CpG-ODNs and pulsed with TRP-2 peptide, were purified by UF-SEC (Ultrafiltration-Size Exclusion Chromatography), validated through DLS, western blot for specific exosome markers, and their proteomic signature was obtained using LC-MS/MS (Liquid Chromatography with tandem mass spectrometry). After passively loading curcumin into EVs, EVs-CURC were co-administered with anti-PD-L1 antibody to a spheroid model comprising murine B16.F10 melanoma cells, DC2.4 dendritic cells and CD8⁺ lymphocytes embedded in 1% commercial ECM. Our results show that the nanoformulation decreases the viability of cells inside the spheroid by 50% compared to control after 48 h (p < 0.0001), leading to a 2-fold reduction of its volume (p < 0.01). Compared to non-encapsulated curcumin, EVs-CURC had 3-fold higher uptake inside the threedimensional model (p = 0.0144). Using the intrinsic potential of extracellular vesicles as drug delivery systems, and the immunomodulatory effects of curcumin, this nano-formulation succesfully enhances the efficacy of the current anti-PD-L1 therapy in melanoma treatment.

Keywords: Curcumin, Extracellular Vesicles, Melanoma

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Comparative analysis of hibiscus and aronia anthocyanins: chemical composition and selective antiproliferative effects on melanoma cells

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Plant-derived bioactive compounds, particularly anthocyanins from *Hibiscus sabdariffa* and Aronia melanocarpa, have garnered increasing attention for their potent antioxidant properties and promising anticancer activities in recent studies (Ezcurra-Hualde et al., 2025). This investigation sought to comprehensively characterize the anthocyanin profiles of these plant extracts and evaluate their differential effects on human melanoma (A375) and non-malignant keratinocyte (HaCaT) cell lines. Anthocyanin-rich extracts were prepared using acidified methanol (0.1% HCl, v/v) extraction methodology. Phytochemical profiling was conducted via reverse-phase HPLC with photodiode array detection (520 nm). Quantitative analysis of total phenolic content employed the Folin-Ciocalteu method with results expressed as gallic acid equivalents (GAE). Total flavonoid content was determined using the aluminum chloride colorimetric assay, reporting results as quercetin equivalents (QE). Antioxidant capacity was assessed through DPPH and ABTS radical scavenging assays, with results standardized to Trolox equivalents (Ezcurra-Hualde et al., 2025). Cellular responses were evaluated via MTT proliferation assay following treatment with extract concentrations ranging from 10-200 µg/mL over 24 hours. Chromatographic analysis revealed distinct anthocyanin signatures, with delphinidin-3-sambubioside predominating in Hibiscus extract and cyanidin-3-glucoside in aronia extract. Aronia extract exhibited superior radical scavenging capacity in both DPPH and ABTS assays compared to hibiscus extract. In cytotoxicity evaluations, aronia extract demonstrated more potent antiproliferative activity against A375 melanoma cells than hibiscus extract. Significantly, both extracts exhibited markedly reduced cytotoxicity toward HaCaT keratinocytes, with aronia extract showing a higher selectivity index than hibiscus extract. These findings demonstrate that hibiscus and aronia extracts possess distinctive anthocyanin profiles and exhibit preferential cytotoxicity toward melanoma cells while substantially preserving non-malignant keratinocyte viability. The observed selective antiproliferative activity merits further investigation into underlying molecular mechanisms and potential applications in melanoma therapeutic strategies

Keywords: anthocyanins; aronia; hibiscus; melanoma; selectivity

Functional study of PAP3/pTAC10 in the Plastid-Encoded RNA Polymerase during chloroplast biogenesis

Calistru Leonard^{\Box_1}, Dorina Podar¹, and Robert Blanvillain²

Plastids, unique to algae and plants, originated from an ancient endosymbiotic event involving a cyanobacterium and a eukaryotic host cell. In angiosperms, plastid gene expression is regulated by two types of RNA polymerases: the nuclear-encoded RNA polymerase (NEP) and the plastid-encoded RNA polymerase (PEP). The latter, existing in two forms—PEP-B and PEP-A—is responsible for transcribing photosynthesis-related genes during chloroplast development. PEP-A is assembled with several nuclearencoded PEP-associated proteins (PAPs), among which PAP3 plays a significant role. This study focused on investigating the localization and potential functional interactions of PAP3 within the PEP complex. Mutants lacking any PAP display an albino phenotype, characterized by a failure to green and subsequent seedling lethality. This phenotype could be further classified into two categories, based on the type of PAP that is lacking. PAPs that are dually localized to the chloroplast and nucleus are involved in phytochrome B (PHYB)-mediated light signaling and play a role in the formation of photobodies. These structures, found in the nucleus, exist in two forms: early photobodies, which are small and numerous, and late photobodies, which are larger and fewer in number. In contrast, PAPs localized exclusively to the chloroplast are not involved in either process. To test these hypotheses, Arabidopsis thaliana pap3 mutants were grown under far-red/red light for 5 days. The results indicated that, unlike PAP8, PAP3 is not essential for PHYB-mediated photomorphogenesis, as mutant seedlings undergo normal deetiolation. Moreover, the formation of photobodies resembled the wild-type phenotype, suggesting that the formation of late photobodies was not hindered. These results are evidence towards the two categories of albino phenotype based on the pap mutant. To investigate subcellular localization and interactions, PAP3 was tagged with fluorescent and epitope tags. GFP-tagged PAP3 constructs, driven by tissue-specific or constitutive promoters, were introduced into onion cells via biolistic and into Arabidopsis thaliang pap3 mutants via Agrobacterium tumefaciens-mediated transient expression. These preliminary transformation assays showed promising GFP signals, supporting the continuation of localization and co-expression studies. This project lays the groundwork for elucidating the role of PAP3 in chloroplast biogenesis and PEP complex assembly. Future work will focus on confirming PAP3 localization, identifying interacting partners, and evaluating functional complementation in pap3 mutant lines to better understand PAP3's contribution to plastid gene expression.

Keywords: chloroplast biogenesis; PAP3/pTAC10; PEP-associated proteins (PAPs); plastidencoded RNA polymerase (PEP).

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Title: Rhizosphere soil microbial communities exhibit resilience over a wide mercury (Hg) contamination gradient

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Mercury (Hg) contamination poses a serious threat to both ecosystem and human health. Soil microbiota, especially those associated with plant rhizospheres, are central to ecosystem resilience, yet their responses to Hg stress remain insufficiently characterized. This study investigates bacterial and fungal communities in the rhizosphere of *Lotus tenuis*—a Hg-tolerant legume—along a Hg soil contamination gradient (40–1964 mg kg⁻¹), by comparing (i) rhizosphere vs. bulk soils within Hg-contaminated sites, and (ii) rhizosphere soils from Hg-contaminated vs. uncontaminated control sites. Microbial community structure differed significantly between control and Hg-contaminated soils. However, alpha diversity remained stable along the Hg gradient, supporting the stress-gradient hypothesis, whereby facilitative interactions maintain microbial function under abiotic stress. In the rhizosphere, Mesorhizobium sp. was abundant across all conditions, consistent with its symbiotic role in nodulation, *Pseudomonas* sp. was notably enriched in contaminated soils, especially within the rhizosphere, suggesting both Hg resistance and a potential non-rhizobial symbiotic function. The merA gene, encoding mercuric reductase, was absent in control soils but detected in 85% of Hg rhizosphere samples, indicating microbial adaptation via active Hg detoxification. Fungal communities were also influenced by Hg, with Glomerales (Mucoromycota)—notably *Rhizophagus*—tripling in relative abundance in contaminated rhizospheres, likely supporting plant water and nutrient uptake. Differential abundance analysis revealed microbial taxa with potential functional roles in Hg-exposed environments: biocontrol agents (Streptomyces), plantgrowth-promoting rhizobia (e.g., Allorhizobium–Neorhizobium–Pararhizobium–Rhizobium, Shinella), and metal- or drought-tolerant genera (Nocardioides, Skermanella). Fungal taxa included dark septate endophytes (Darksidea, Acrocalymma paeoniae), growth-promoting fungi (Mortierella alpina, *Chaetosphaeronema*), plant pathogens (*Aspergillus* spp.), and potential pathogen suppressors (*Humicola*, *Vishniacozyma*). These results enhance our understanding of rhizosphere microbiome adaptation under Hg stress and identify promising microbial partners for Hg phytoremediation strategies.

Keywords: *Lotus tenuis; merA*; mercury; phytoremediation; pollution.

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Maximizing microbial DNA yields: practical solutions for sampling amphibian skin microbiome

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The skin microbiome of amphibians plays a crucial role in protecting the host against pathogenic infections. Understanding the microbial diversity associated with amphibian skin is therefore essential. However, the challenge of effectively recovering microbial biomass from amphibian skin remains underexplored. This study aims to evaluate the feasibility of different swab types and DNA extraction kits for maximizing microbial DNA recovery. We sampled the skin microbiome of Triturus vulgaris ampelensis (n=33), captured alive from a pond located in Făget Forest (Clui-Napoca, Romania). Each individual was rinsed with sterile distilled water to remove transient microbes and swabbed for 30 seconds on the dorsal, ventral and lateral parts of the body. Three different swabs were used in this study: cotton, foam, and flocked swabs (dry swabs and eNAT system). Each individual was swabbed with one particular type of swab. DNA was extracted in the laboratory using three commercial kits: (a) ZymoBIOMICS DNA Miniprep Kit, (b) Ouick-DNA HMW MagBead Kit (both from Zymo Research, USA), and (c) DNeasy PowerSoil Kit (Qiagen, Germany). DNA quantification was performed using the Oubit 4 Fluorometer (Invitrogen, USA), and samples were submitted to Novogene (Germany) for 16S rRNA gene amplicon sequencing. The highest DNA yield $(13 \ \mu g \pm 1.9 \ \mu g)$ was achieved using the a) kit in combination with the flocked swabs (eNAT system), followed by the pairing of the same kit with flocked dry swabs ($4 \mu g \pm 0.21 \mu g$). The flocked swabs (eNAT system) also performed well when used with the b) kit (1.9 μ g ± 0.069 μ g). In contrast, cotton and foam swabs yielded lower DNA quantities, though still adequate for applications such as amplicon sequencing. Notably, the PowerSoil kit was only effective when used with flocked dry swabs and proved less efficient with flocked (eNAT system), cotton, or foam swabs. These results suggest that selecting the right swab and extraction kit is critical for maximizing microbial DNA recovery in amphibian microbiome studies.

Keywords: Amphibian skin, DNA extraction, microbiome, swab types.

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Testing the germination and reactivity of *Angraecum caleolus* using *in vitro* cultures

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In vitro germination of orchids seeds is a rather difficult process, due to the symbioses that these plants develop with various fungi. *Angraecum calceolus* Thours is an orchid that has a fairly limited range, naturally occurring only in Mozambique and Madagascar, Comoros, Mascarene, Seychelles islands. It grows in forests at altitudes between 100-1 400 m, in areas with moderate rainfall. It is a small, lithophytic or epiphytic orchid, with rapid growth. Our study aimed at testing the seed germination by *in vitro* culture and the reactivity of the species on different culture media, in order to obtain an optimal variant for micropropagation and acclimatization of seedlings to *ex vitro* conditions. Thus, 6 variants of culture media were tested for multiplication, having as basal medium MS supplemented with 0.5 and 1 mgl⁻¹ benzylaminopurine (BA) and 0.5 and 1 mgl⁻¹ kinetin (K). The MS variant without hormones was the control and we also tested the MS ½ variant. The maximum number of shoots was obtained on the MS variant + 1 mgl⁻¹ BA, and the highest shoots were obtained on the MS variant without hormones. The plants were acclimatized on *Sphagnum* with activated carbon in covered pots, from which the lid was gradually removed. After acclimatization in the vegetation chamber, the plants were transferred to the greenhouse.

Keywords: Angraecum caleolus; in vitro propagation; orchids.

Earth sciences and ecosystem dynamics across temporal scales

KEYNOTE

Cave deposits as archives of past climate and environmental change

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Cave deposits offer exceptional archives for reconstructing past climate, environmental changes, sea-level fluctuations, and even human activity. This presentation highlights results from my research on speleothems, cave ice, and guano deposits across a wide range of settings—including Romania, Spain, Italy, Iceland, Greenland, and the USA—demonstrating their value in tracking Earth system processes across diverse timescales. Speleothems, through U-Th dating and stable isotope analyses, provide high-resolution records of hydroclimatic variability and vegetation dynamics. Examples from Romania and Mallorca illustrate how stalagmites capture regional responses to hemispheric-scale climate events and glacialinterglacial transitions. In coastal karst environments such as the Mediterranean regions, Yucatan, and Tahiti, speleothems and associated marine notches and terraces also serve as proxies for relative sea-level reconstructions. By combining precise elevation measurements and geochronology, we can infer past sea-level positions and tectonic uplift rates, offering insights into ice sheet stability and global sea-level budgets from Miocene to present. Cave ice deposits, such as those in Scărisoara Ice Cave (Romania), preserve stratified records of temperature, source of precipitation, and atmospheric composition over the Holocene, while guano accumulations, through their isotopic and elemental signatures, provide valuable data on past ecosystem shifts, trophic structure, and anthropogenic impacts. Importantly, many of these archives also preserve evidence of human presence and activity within caves—ranging from Neanderthal footprints to traces and cultural material embedded in ice. Together, these diverse deposits form a powerful multi-proxy toolkit. By integrating mineralogical, geochemical, and isotopic analyses, and situating results within regional tectonic and climatic frameworks, we can better understand the complexity of Earth's climate system. This presentation emphasizes the importance of cross-disciplinary collaboration and continued methodological innovation to unlock the full potential of cave environments as paleoclimate archives.

Keywords: guano; ice; minerals; speleothems; sea level.

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The hydroclimate of Mlaca Tătarilor (Central Romania) in the last two millennia

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The present study analyses a peat sequence from the Mlaca Tătarilor peat bog (Arpașul de Sus, Southern Transylvania) to reconstruct local environmental and hydroclimate changes over the past two millennia. Using peat physical properties (radiocarbon dating, lithology, loss on ignition, and magnetic susceptibility), we reconstructed the palaeoenvironmental phases of the basin. Pollen analysis was used to reconstruct the vegetation history and assess human impact, while testate amoebae analysis allowed us to infer past water-table depth in the peat bog. Over the past 500 years, an increase in anthropogenic impact and a notable drying trend in the peat have been observed. However, a peat restoration campaign conducted in 2016 has already yielded observable outcomes in our results: a significant decrease in the water-table depth, as indicated by testate amoebae communities, suggesting wetter conditions in the peatland, which may facilitate peat growth.

Keywords: hydroclimate, peatland, peat restoration, testate amoebae, water-table depth

Mineralogy and geochemistry of carbonates containing rich *Frutexites-microcoprolites* assemblages from Mesozoic marine cryptic cavities (Romanian Carpathians)

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Frutexites is a challenging microstructure commonly found in carbonate rocks. The genus Frutexites was defined by Maslov (1960) to describe submillimeter-sized microfossils characterized by a generally dendritic shape composed of divergently branched micro-columns. These microstructures manifest as iron-rich and/or manganese-rich minerals, and/or carbonate-rich, and/or phosphate-rich minerals. The genesis of *Frutexites* remains the subject of numerous debates, with discussions involving various processes, including abiotic, biotic, or coupled abiotic-biotic factors. While Frutexites-like microstructures have been documented in marine environments throughout the Proterozoic, Palaeozoic, and Mesozoic deposits, recent occurrences of Frutexites-like microstructures have only been reported from various nonmarine environments. Our study highlights peculiar occurrences of *Frutexites* alongside a high concentration of microcoprolites produced by decapod arthropods from ancient marine cavities and fractures opened in Middle Jurassic and Upper Triassic carbonate rocks of the Romanian Carpathians. The mineralogical composition of the studied *Frutexites* and microcoprolites is characterized by goethite and calcite (for the Jurassic occurrence) and hematite, calcite, and scarce Fe-Mn oxides and phosphates (for the Triassic occurrence). Hematite, Fe-Mn oxides, and phosphates appear to be associated systematically with microcoprolites and burrows and with Frutexites-like microstructures. Nonetheless, these minerals are also found dispersed throughout the sediment matrix. Geochemical proxies, including the distributions of rare earth elements and yttrium (REE+Y), indicate largely normal marine conditions, with limited terrigenous input. However, negative cerium anomalies and positive lanthanum anomalies indicate conditions slightly different than modern seawater. Moreover, the apparent cyclic variation of yttrium anomalies, from positive to negative, is likely to represent variations in oxygenation levels of this particular environment. The carbon stable isotope composition of the sediment containing these Frutexites-like microstructures does not show any significant shift from the normal marine composition. The abundance of microcoprolites and numerous burrows with variable dimensions suggests that the crustacean arthropods constituted the autochthonous biota of the studied cavities. The development of *Frutexites*-like microstructures occurred in these cryptic, aphotic environments during intervals of very slow sedimentation under dysoxic or dysoxic-anoxic conditions, when the abundance of organic matter-rich microcoprolites facilitated intense bacterial activity within the fecal pellets.

Keywords: Frutexites, Geochemistry, Marine carbonates, Microcoprolites, Carpathians

Environmental mineralogy studies on some built structures in Cluj area

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Construction materials used in various built structures and modern infrastructure are subject to degradation processes caused by physical, chemical, and environmental factors. This study investigates the mineralogical behavior and petrographic changes in samples collected from representative structures in Cluj area. Representative samples were collected from several locations (eg. Central Building of Babes Bolyai University, the Piarist Church, the Memorandists Monument, the RomTelekom building), and the Tureni and Lita. All samples were the subject of mineralogical and petrographical investigation using optical polarized microscopy and x-Ray diffracometry. Different degradation processes were identified as dissolutions, damage caused by freeze-thaw cycles, carbonatation, sulfate attack etc. some of them with specific secondary mineral phases. While the physical processes as dissolution of calcite (Piarist Church) and freeze-thaw cycle (plaster of the inner yard of Babes Bolyai University) led mainly to mechanical disintegration, the other processes led to formation of specific minerals as secondary calcite, portlandite, melanterite, rozenite, szomolnokite, ettringite, gypsum, thenardite, brushite, illite, iron oxi-hydroxiedes etc. as the result of interaction between natural or syntetic components from the raw materials and external factors such as temperature, humidity, or infiltration of mineral-rich solutions. Chemical alteration of natural or syntetic row materials in construction reveals the key role of secondary minerals in their degradation. Sulfate mineral precipitation (e.g., melanterite, rozenite, szomolnokite, gypsum) and carbonation are the main processes, both reducing durability. Sulfate minerals are manly generated as the result of metal sulfides (eg. pyrite and marcasite) and local conditions, while carbonation, driven by the interaction between primary calcite and CO₂, leads to formation of secondary calcite, especially in areas with water infiltration. Sulfate attack, as seen in UBB concrete, produces ettringite and gypsum, causing damaging expansion. To prevent these effects, it's crucial to use quality-controlled, sulfide-free aggregates, optimize mixtures, prevent water infiltration, and regularly monitor and maintain structures.

Keywords: Cluj area, degradation processes, environment, mineralogy

Dung fungal spores in modern samples as indicators of herbivore grazing pressure

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Herbivores shape vegetation dynamics by maintaining more open landscapes. Coprophilous fungal spores (CFS) are widely used to reconstruct past grazing activity, yet their potential for assessing grazing pressure remains underexplored. Here, we analyse CFS composition in surface soil and dung samples across diverse habitats in Romania, representing the first assessment of their diversity and environmental drivers in European lowland agropastoral ecosystems. We identified seven CFS types, with Sordaria, Podospora, Sporormiella, and Delitschia emerging as the most reliable indicators of grazing pressure, whereas Arnium, Bombardiaceae, and Trichodelitschia were present less commonly. The co-occurrence of fungal spore types with occasional coprophilous affinities such as Apiosordaria, Cercophora, Chaetomium, Coniochaeta, Gelasinospora, and Neurospora, can also reflect grazing activity. Glomus proved a strong indicator of heavily grazed environments. We observed a higher abundance of *Podospora* on cattle dung, Sporormiella on sheep dung, and Sordaria on both cattle and sheep. CFS abundance also increased in moist habitats, spring samples with higher moisture and fresh biomass, and areas where animals drink and rest. Overall, CFS abundance closely tracked grazing pressure, though quantitative herbivore density reconstructions remain challenging due to animal diversity, mobility, and spore dispersal. Trends in dung spore concentrations mirrored percentage data but captured abundance more effectively at lower CFS counts. This study highlights avenues for further research into herbivore grazing intensity and ecosystem dynamics in palaeoecological records. One of the key suggestions is to broaden the range of fungal spore types used to reconstruct past grazing intensity, along with the need to standardize identification protocols to enhance comparability across studies.

Keywords: agropastoral ecosystem; coprophilous; grazing activity; herbivore density; spore dispersal

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Alluvial gold from the Eastern and Southern Carpathians, Romania

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Romania has been recognized as a significant source of gold since at least the Roman era, with total gold production estimated between 1,500 and 2,000 tons, according to Cook and Ciobanu (2004). The primary gold-producing regions include the South Apuseni Mountains, particularly the "Golden Quadrilateral" area as named by Ghitulescu and Socolescu (1941), as well as the Baia Mare ore district located in the northwestern segment of the Eastern Carpathians. In addition to the primary gold deposits. Romania is home to five known placer gold deposits: Aries Valley, Cibin-Olt Valley, Nera/Bozovici, Pianu and Râureni, Among these, Pianu stands out as the most notable, with well-preserved evidence of placer gold mining dating back to the 2nd and 3rd centuries AD. This study focuses on two alluvial gold occurrences within the Eastern and Southern Carpathians. The first occurrence is situated along the Cheia and Olănesti River, which originates in the Căpătânii Mountains of the Southern Carpathians and flows southward. The second occurrence is located on the Bogata River at the border of the Eastern Carpathians with the Moldavian Platform, flowing west to east. The gold grains from the Southern Carpathian occurrence reach a maximum size of 4 mm, while those from the Eastern Carpathian occurrence reach up to 6 mm. In both cases, the grains predominantly exhibit discoidal to subdiscoidal shapes, with roundness varying from rounded to intermediate. A notable feature is the presence of quartz still attached to the gold grains at both locations, indicating a short transport distance. Both occurrences are considered to have originated from orogenic gold mineralizations, as indicated by chemical analyses carried out at the CAMECA SXFive device of the Geology Department of the Babes-Bolyai University. The gold source in the Southern Carpathians occurrence is likely the Valea lui Stan and/or Costesti gold-bearing ore deposits. In contrast, the source of the gold in the Eastern Carpathians occurrence remains for the moment unknown.

Keywords: alluvial gold, EPMA, placer mining

Posters

The anthropogenic impact on diurnal butterfly communities *(Lepidoptera)*: An insight from three Natura 2000 sites located in Brașov county

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Insects support all biodiversity and sustain the good functioning of the habitats and ecosystems, being the key to survival for many other species. It is well known that butterflies are good bioindicators and pollinators. By studying Lepidoptera order one can assess the quality of an ecosystem or habitat, based on the sensitivity of some species towards changes caused by different factors. The objective of this study is to determine whether anthropogenic impact affects or not the diurnal butterfly communities from 3 sites of Natura 2000 network, located in Brasov county. Anthropogenic impact comes in various forms, but the main impacts outlined in this study are: intensive agricultural practices (e.g. livestock grazing) and urbanization. The linear transect method was used to observe and identify the individuals along 9 selected transects, 3 for each site. Based on the data set collected on the field during the flying? season, several statistical tests were conducted. And so, one of the hypotheses of the study was validated and sustained by the results. Some diversity indices (e.g. species richness, abundance) relevant for the study were less-represented regarding the first site, in comparison with the other two sites. It can be summarized that the butterfly diversity of the first site is possibly more affected by intensive grazing than the other two sites. In the case of the second and the third site, the pastures are not enclosed by coniferous forests and the protected area management is more accessible: here the human activities consist in keeping the grasslands composed of native species that sustain the diversity of pollinators, and cultivating some small crop areas. Therefore, this short-termed study can only highlight the importance of undergoing more detailed research in Brasov county, in order to establish the dynamic of butterfly diversity, along with proper solutions of conservation.

Keywords: anthropogenic impact, biodiversity, diurnal butterflies, grazing, protected areas

Shedding lights into underground diversity: groundwater harpacticoids (Crustacea. Copepoda) from Romania

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Harpacticoids are copeped crustaceans dwelling diverse habitats, including groundwater where they exhibit unique morphologic adaptations, niche segregation and habitat specialization. Among other groundwater taxa, harpacticoids are model organisms due to their high level of speciation and endemicity at small spatial scale. In Romania, the harpacticoids have been studied from the first half of the 20th century by the Swiss researcher P. A. Chappuis. Later on, the investigations have continued by A. Damian-Georgescu and punctually L. Botosaneanu, E. Serban and D. Zincenco. We here aim to provide a general overview of harpacticoid diversity and distribution from groundwater habitats at the country level, using geographic information systems (GIS) of the record sites available to date. Based on the constructed database we further summarise the information on species ecology, genera diversity, and stygobites distribution in distinct karst areas identifying the gaps in harpacticoids knowledge. Currently, there are 60 species and subspecies of harpacticoids from over 200 records in Romanian groundwater habitats. The species number is extremely low considering the total surface of the country (256.000 km²). Almost 70% of the species are stygobites, many of them with restricted distribution to a single massif. A high speciation has been observed within *Elaphoidella* and Parastenocaris, all species being exclusively stygobitic. The striking distribution within stygobite species across distinct massifs, such is for example *Elaphoidella putealis*, present in both Apuseni and Banat karst, suggests the need for more sampling in both areas corroborated with genomic sequencing, to confirm the records in such remote locations. The distribution, taxonomy and ecology of groundwater harpacticoids are open subjects that create the potential for further research and exploit their potential to be used as model group taxa to interpret the factors shaping their distribution at local and in a regional context.

Keywords: aquifers; distribution; diversity; endemicity; stygobitic

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Intraspecific interactions within the population of *Anas platyrhynchos* on Lake Chios in Cluj-Napoca

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Anthropization has become a significant global issue in today's world. The expansion of anthropogenic areas affects interactions between individuals of waterfowl species. Their adaptation to urban conditions includes behavioral changes, including intraspecific interactions. Understanding these changes is essential for the effective conservation of species and minimizing the adverse effects of anthropogenic activities. In this context, the study focused on the quantitative and qualitative of intraspecific interactions within the Anas platyrhynchos population inhabiting Lake Chios, Cluj-Napoca. Observations were carried out from November 2023 to October 2024, during a total of 32 field trips. Individual interactions were monitored and documented in an ethogram over a one-hour observation period. Interactions were grouped into three behavioral categories: aggression, care and courtship. Population dynamics, sex ratio and social structure (solitary individuals, pairs or groups) were also monitored. From the observations, it was found that aggressive behaviors were the most frequent (n=87), accounting 52% of the total recorded interactions (n=166). The rarest behavior were the courtship interactions (n=30), representing 18%. Most interactions between males and females were courtship. Qualitatively, the number of specific interactions varied by month, with most recorded in April (n=32) and fewest in November (n=3). It can be concluded that the dynamics of intraspecific interactions are closely linked to both natural biological cycles and humaninduced disturbances, with human presence being a key factor.

Keywords: Anas platyrhynchos, anthropization, behaviors, interactions, observations

Preserving Heritage: the revision of the zoological collection held by Andrei Mureșanu High School Natural History Museum, Dej, Romania

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The Andrei Mureşanu National College in Dej houses an impressive natural science collection, with origins dating back to the school's founding in 1897. Over the decades, the collection expanded through a variety of sources, becoming one of the most significant of its kind in the region. Despite containing numerous specimens of notable faunistic and scientific-historical value, the collection had not been thoroughly documented, its provenance clarified, or its condition improved — until now. As part of our conservation-focused project, we conducted a comprehensive survey of the collection, re-identified species, and carried out preservation work. We also consulted historical written records to trace the origins of individual specimens. In total, we identified 450 zoological specimens, the majority of which are vertebrates — particularly birds. The collection spans the period from 1897 to 1999, with most specimens originating from the Dej region, although it also includes many exotic species. In our presentation, we will provide a detailed overview of the collection, with special emphasis on its faunistic and scientific-historical significance.

Keywords: collection; Dej; museum; zoology

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Integrating classical and molecular tools for biodiversity monitoring in subterranean karst environments

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Subterranean karst ecosystems (including caves and springs) have high species diversity with specialised physiological and morphological adaptations. Due to the subterranean environment's complexity and heterogeneity, along with species identification challenges, estimating biodiversity and monitoring species remain difficult. The lack of taxonomists for several groups in recent decades has hindered the development of up-to-date country-level databases and limited collaboration across European regions. Within the Sub-BioMon project, we aim to develop and test a standardised monitoring protocol to assess the biodiversity of selected subterranean invertebrates, crustaceans, beetles, and spiders across karst areas from six European countries. While some countries have national cave or species monitoring protocols, a unified monitoring method for rare subterranean species is still lacking. Combining classical taxonomy with environmental DNA (eDNA) - a useful tool when direct observations or specimen collection is difficult - we will monitor the biodiversity in a selected number of Romanian caves and springs, over one year. Our compiled national database includes over 1000 records from 163 species, including 190 troglobites (165 beetles and 25 spiders) and 108 stygobites (41 species of harpacticoids, 38 amphipods, 18 podocopids and cyclopoids, eight Bathynellacea and three Isopoda). However, the DNA database contains sequences for only 21 species, highlighting the lack of molecular studies on subterranean fauna. This project promotes eDNA as a tool to overcome the "taxonomic barrier", enhancing species identification through traditional morphology and helping biodiversity assessment. The proposed standardized protocol aims to strengthen the conservation strategies, providing valuable data for stakeholders managing karst areas nationally and across Europe. It also supports the conservation goals listed in the EU Habitat Directive, 8310 "Caves not open to the public". This approach will serve as a scientific foundation for longterm subterranean biodiversity monitoring contributing to the protection of one of the most fragile and less studied ecosystems.

Keywords: conservation, eDNA, monitoring, karst, biodiversity

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Digitizing entomological collections: scientific valorization of the Vasile Vicol Carabidae collection

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The Carabidae (Coleoptera) collection realized by Professor Vasile Vicol and deposited at the Zoological Museum of Babes-Bolyai University constitutes a valuable entomological archive for the study of beetle biodiversity in Romania, with a particular emphasis on the region of Transylvania. Comprising 1,392 specimens representing 120 species, the collection includes both native and exotic taxa, enriched through international specimen exchanges. High species richness is recorded in counties such as Mureş, Sălaj, Cluj, and Sibiu, with the locality of Panic (Sălaj) being especially well represented. Assembled primarily in the latter half of the 20th century, the collection reflects Professor Vicol's extensive fieldwork and collaborations with fellow entomologists during a period marked by heightened faunistic research in Romania. This era contributed substantially to the consolidation of regional biodiversity records through active documentation and specimen exchange. The digitization process involved the systematic reorganization of specimens, standardized labelling, and the development of a comprehensive database. Taxonomic verification presented several challenges, particularly in the case of specimens with uncertain or historically incorrect identifications. The scientific valorization of museum beetle collections is essential for advancing taxonomic and biogeographical research. Furthermore, digitized data support the reconstruction of historical species distributions and provide critical insights for ecological and conservation studies. Integration into open-access platforms such as GBIF facilitates the correction of taxonomic inaccuracies and ensures broader accessibility to biodiversity information. This case study underscores the enduring scientific relevance of legacy entomological collections and highlights the importance of their preservation, digitization, and incorporation into global biodiversity information systems.

Keywords: Carabidae, Coleoptera, digitization, museum collections

Tracing pig diets in 5th-6th century Transylvania: a stable isotope approach

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The aim of this study is to generate insights into pig diet from the 5th to 6th century in Transylvania, more specifically from the fortress of Legio V Macedonica at Potaissa (Turda, Cluj County). The samples, archaeologically dated to the Early Migration Period, consist of six pig bones that were analyzed using stable carbon and nitrogen isotopes from bone collagen. The bone collagen was extracted using a modified Longin method followed by an ultrafiltration step. The stable carbon isotopic signatures are indicative of a predominantly C3 plant-based diet, while the nitrogen values reflect little to no added animal protein in the diet of the pigs, findings that are consistent with established knowledge for this period. The significance of this study lies in the novelty of its isotopic data for this region and time frame, contributing to a more cohesive understanding of swine diet and domestic animal feeding practices.

Keywords: Bone collagen, carbon, nitrogen, pig diet, stable isotopes

Funding: Doctoral research grant

Micropropagation of the critically endangered *Convolvulus persicus*: A step towards ex situ conservation

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In this study, we present the micropropagation and acclimatization of *Convolvulus persicus* L., a rare species from Romania. Our goal was to obtain as many plants as possible for the acclimatization phase, knowing that a significant number of plantlets are lost during this stage. Seeds were used to initiate in vitro culture. The seeds were sterilized and inoculated onto MS medium. Of the 20 seeds inoculated in the medium, only one germinated, serving as the source of plant material. From the in vitro germinated seed, explants were subsequently used for multiplication and acclimatization. For multiplication, we used six variants of MS culture medium supplemented with K and BA in different concentrations, with 18 explants per variant. The best multiplication and the tallest plants were obtained on MS medium without hormones. Regarding the two hormones used, the highest multiplication rate was observed on MS medium supplemented with 0.5 mg/L BA, while the tallest plants were obtained on MS medium supplemented with 0.5 mg/L K. After multiplication, the plants were transferred for acclimatization using the "floating perlite" system on distilled water and $\frac{1}{2}$ MS, in a container covered with plastic wrap, which was gradually removed. After acclimatization, the plants will be transferred to the botanical garden collections, where they will be monitored for seed The use of in vitro multiplication for the ex situ conservation of rare and production. endangered species with low seed production and predominantly vegetative propagation, such as *Convolvulus persicus*, can be beneficial due to the ability to obtain a large number of plantlets in a relatively short time. These plants, after acclimatization, can be used for ecological restoration programs in affected habitats.

Keywords: conservation; *Convolvulus persicus;* ex situ; micropropagation.

The diversity of herpetofauna from the sites ROSCI0420 Oprănești and ROSCI0432 Prunișor

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This study focuses on assessing amphibian and reptile species within the ROSCI0420 Oprănești and ROSCI0432 Prunișor sites, both situated in Mehedinți County. These areas stand out due to their unique climatic traits, which support the county's rich herpetofaunal diversity. Conducted between March and September 2024, the study aligns with the primary activity period for reptiles and amphibians. Species identification was carried out using transects along designated routes, and the resulting data were further analyzed. Seven species of reptiles and four species of a dominant species like *Lacerta viridis* in the sites but also viable populations of pretty rare species like *Podarcis tauricus*. This research is scientifically significant, confirming the presence of key herpetofauna species in Mehedinți County and providing a valuable foundation for future ecological studies in the region.

Keywords: assessment; population; herpetofauna; Mehedinți

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Cryptic cave spiders: is *Carpathonesticus plesai* a separate species from *Carpathonesticus biroi*?

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Spiders are one of the most abundant cave invertebrates, playing a major ecological role in caves trophic chain as they are known as top predators. In Romania, there are approximately 74 species of troglophile (63, including subtroglophiles) and troglobiont (11) spiders. This number is highly underestimated when compared to other karst areas from southeastern Europe. Several populations with close distributional range might be cryptic species of spiders, a very common phenomenon in caves and an important component of biodiversity. Carpathonesticus Lehtinen & Saaristo, 1980 is one of the most specious genera in the Romanian Carpathians, with 21 species. Carpathonesticus biroi (Kulczyński, 1895) and Carpathonesticus plesai (Dumitrescu, 1980) are two eutroglophile species of spiders, endemic to the Apuseni Mountains, in the northwestern Romania. Both species have close distributional ranges and show pronounced phenotypic similarities. Their current morphology provides few distinctive morphological characters and limited reliable diagnostic information. Here, we aim to investigate the morphological differences among specimens of *C. plesai* and *C. biroi* coupled with molecular analyses of sequences available in GenBank. The currently known geographic distribution of both species is also illustrated using GIS. Our work highlights the needs to improve the taxonomic descriptions of cave spiders with minute morphological characters coupled with interspecific and populational molecular investigations. This will help to validate the potential cryptic species and test taxonomic validity in very closely related species. This will also improve our efforts to adopt specific conservation strategies to protect the biodiversity of cave spiders in particular and of cave invertebrate biodiversity in general.

Keywords: Araneae; Carpathonesticus; Nesticidae; taxonomy.

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Digitizing to preserve: the scientific value of the Vicol Macrolepidoptera collection in biodiversity conservation

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The Zoological Museum of Babeş-Bolyai University in Cluj-Napoca houses a valuable collection of moths (Macrolepidoptera), assembled between 1975 and 2001 by Professor Vasile Vicol from Târgu Mureş. The collection comprises 3,247 specimens, representing 491 species collected from various regions of Romania. This collection reflects a remarkable diversity of species, including numerous rare or protected taxa, and serves as a significant scientific resource for the study of Lepidoptera biodiversity in Mureş County and, more broadly, throughout Romania. The study of the collection provides insights into species' habitats, geographical distribution, ecological preferences, and conservation status as listed in Romania's Red List of Lepidoptera. The collection includes several rare or protected species (Natura 2000 species), such as *Eriogaster catax, Proserpinus proserpina, Conisania poelli ostrogovich, Drasteria caucasica, Watsonarctia deserta*, and *Periphanes cora*. Many of these were collected from ecologically sensitive areas, such as the hills and floodplains around the village of Sânmărghita.

Museum insect collections play a crucial role in biodiversity conservation by providing essential data for taxonomic, biogeographical, and ecological studies, as well as reference points for monitoring environmental change. They act as biological archives that document historical species distributions and help identify trends in population decline or expansion, thus supporting evidence-based conservation policy. Preserving this collection and integrating it into the scientific domain not only honors Professor Vicol's legacy but also contributes significantly to the documentation and protection of Romania's entomological biodiversity.

Keywords: digitization, Lepidoptera, museum collections, Natura 2000,

Life on the river bottom: the status of benthic invertebrates from the Someșul Mic in the city of Cluj-Napoca (Transylvania, Romania)

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The aim of this study was to analyze the structure of benthic macroinvertebrate communities in the Somesul Mic River, including two tributaries and one canal located within the urban area of Cluj-Napoca, Romania. A total of 10 sampling sites were selected: six on the main river channel, one on the Valea Gârbăului tributary, two on the Morii Canal, and one on the Nadăs tributary. Biological samples were collected on October 18 and October 21, 2024, targeting benthic invertebrates, with *in situ* measurements of key physico-chemical parameters performed at each site. In total, 21 taxa were identified at various taxonomic levels, including representatives of the sensitive EPT groups (Ephemeroptera, Plecoptera and Trichoptera). Principal Component Analysis (PCA) was performed to explore the community structure and the environmental parameters. The results indicated that the tributaries and the Morii Canal were dominated by pollution-tolerant taxa (e.g. Chironomidae, Oligochaeta, Isopoda), suggesting reduced water quality. In contrast, higher proportions of EPT taxa were recorded at sites on the main river channel, indicating better ecological conditions. Moreover, subsequent PCA results showed a clear association between tributary and canal sites and elevated conductivity and phosphate levels, highlighting the influence of anthropogenic pressures on benthic community structure. Biological indices were calculated to assess ecological quality, focusing on the relative abundance of tolerant taxa (Oligochaeta and Chironomidae, the OCH index) and sensitive EPT taxa. According to the Extended Biotic Index (EBI), water quality along the main river course ranged from class II (good) to class III (moderate). In contrast, lower water quality was recorded in the tributaries, reaching class IV (poor) at station NAD.

Keywords: abundance, community structure, dominance, physico-chemical parameters, water quality.

Investigation of the effects of mansorin on memory processes in a zebrafish (*Danio rerio*) animal model

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Alzheimer's disease (AD) is a progressive neurodegenerative disorder that affects millions of people worldwide, generating a significant socio-economic impact. The limitations of current treatments and the associated adverse effects drive interest in natural compounds with therapeutic potential. Mansorine (MA), a coumarin derived from *Mansonia gagei*, is recognized for its antioxidant and anti-inflammatory properties. The aim of this study is to investigate the effects of mansorine on memory, using zebrafish (Danio rerio) as a preclinical model for AD. To induce an AD-like amnesia model, okadaic acid (OKA) was administered. Fish were divided into treatment groups with different doses of MA. Cognitive functions were assessed by two behavioral tests: the Y-maze and the novel object recognition (NOR) test. Locomotor activity and recognition memory were monitored and compared with the control group treated with OKA alone. Mansorine administration produced significant changes in locomotor activity in the Ymaze test, indicating a stimulatory effect on exploratory behavior. In the NOR test, fish treated with MA showed an increased preference for the novel object, suggesting an improvement in recognition memory. These results support the hypothesis regarding the neuroprotective potential of MA in Alzheimer's-type cognitive disorders. Mansorin demonstrates a beneficial effect on memory and locomotor behavior in the *Danio rerio* model, suggesting its potential for future use in therapeutic strategies for AD.

Keywords: Alzheimer's disease, mansorin, Danio rerio, memory, natural compounds.

Assessing spruce forest conservation status in respect to lichen flora in the Apuseni Mountains

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Romania's forests are undergoing rapid degradation, leading to the decline of associated cryptogamic species, which are crucial components of biodiversity. These organisms are highly sensitive to habitat quality and forest management practices. Currently, forest habitat quality is evaluated primarily based on vascular plant indicators. To address this limitation, the present study focused on three forest plots in Cluj County, each characterized by a different management history: an unmanaged (secular) forest, a forest clear-cut 50–100 years ago, and a selectively thinned forest. In this study, we aimed to answer the following questions: i. Are there differences in the lichen flora among the three categories of forests?; ii. Which species could serve as good indicators of the quality of spruce forests?; iii. Which species can be used for monitoring the quality of these forests? Within each 1-ha plot, at least 10 living trees and 10 logs or stumps at various stages of decay were randomly selected to assess the lichen communities. A species database was compiled for each plot, and the results were subsequently compared. The clearcut forest hosted 21 lichen species, while only 13 species were recorded in the thinned forest. Of the 37 species identified across all sites, 29 were found in the secular forest, 14 of which were exclusive to it. Several species, such as Bryoria spp., Usnea hirta and Alectoria sarmentosa, were found exclusive in the secular forest and can be considered indicators of mature, well-developed forest ecosystems.

Keywords: lichen diversity, bioindicators, forest management & conservation, biomonitors

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Salt shock: testing the limits of freshwater phytoplankton

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Freshwater salinization (FS) - accelerated due to global changes including climate, sea level rise. and urbanization -is a growing global threat to key ecosystem services including the provision of water, biodiversity and food security. Despite its importance and potential damage, we are only starting to understand the ecological effects of FS. Phytoplankton - planktonic microalgae and cyanobacteria - dominate primary production and are key regulators of water quality. fisheries, and carbon sequestration. Yet, our understanding of their response to FS is poor, as most studies on FS have focused on invertebrates and fish. Accordingly, we aimed to evaluate the salinity tolerance of four globally abundant species of phytoplankton from two key freshwater phyla (Cyanophyta and Chlorophyta). We predicted that growth rates would decline with salinity, and that there would be strong species-specific differences in tolerance. In replicated microcosm growth assays, we compared specific growth rates of Microcystis, Synechocystis, Desmodesmus and Deuterostichococcus across concentrations of NaCl across a wide range of salinity (0-30 g/L) over a period of 10 days. As expected, growth rates declined consistently and significantly with salinity. We also found a large gradient in salinity tolerance, with Cyanobacteria species being less tolerant than the Chlorophyta species, and the toxin producing Microcystis the most sensitive species. In contrast, Deuterostichococcus (Chlorophyta) continued to grow at salinities up to 30 g/L NaCl (i.e., marine conditions). Overall, we found that while salinity reduced growth significantly, species-specific effects were strong. Hence, the effects of FS can be expected to alter the productivity and community composition of freshwater phytoplankton and species interactions. Our results have implications for water managers and ecologists alike. Identification of salinity tolerant phytoplankton species is important for bioremediation. Moreover, the potential for evolutionary adaptation of phytoplankton has implications for ecosystem resilience in a future world.

Keywords: Chlorophyta; Freshwater salinization; phytoplankton; salinity; tolerance.

Optimizing *Lotus tenuis* Growth and Seed Multiplication for Phytomanagement Applications

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The Fabaceae plant family comprises approximately 18 000 species with various physiological traits, including nitrogen fixation in soil, that enable survival in harsh environments. The legume Lotus tenuis is capable of growing in different degraded soils, including alkaline, acidic, droughtaffected, and heavy-metal-contaminated environments, making it a promising candidate for nature-based restoration strategies. However, knowledge regarding its growth cycle and laboratory propagation methods are scarce in the scientific literature. The aim of this study was to comprehend the life cycle of *L. tenuis* under controlled laboratory conditions to enable successful seed multiplication for use in future physiological and molecular biology experiments. Seeds were collected from four distinct degraded sites: mercury (Hg), hexachlorocyclohexane (HCH), copper (Cu), and saline (NaCl)-affected soils. Following mechanical scarification, seeds were sown in liffy pellets and grown for one month. Seedlings were subsequently transferred in pots and cultivated at 23°C under a long-day photoperiod (16/8 h light/dark), 70% humidity, and light intensities reaching up to 850 μ mol m⁻² s⁻¹. Flowering occurred after 4-5 weeks, and bumblebees (Bombus terrestris) were introduced separately for each genotype to allow natural pollination. After 15 weeks of growth, mature and dry pods containing viable seeds were harvested. The average of production was approximately 200 seeds/plant. This established protocol proved effective for the multiplication of L. tenuis seeds collected from different problematic soils, ensuring their availability for future phytomanagement experiments. To our knowledge, this is the first described growth protocol for this legume species, also providing insights into its life cycle. We further aim to characterize L. tenuis in the context of environmental pollution, particularly mercury. By employing cuttingedge molecular techniques, we will explore how rhizosphere and nodule microbial communities are shaped by distinct stressors, which microorganisms are selectively recruited into the nodules, and what functional benefits provide in terms of plant growth, health, and contaminant reclamation.

Keywords: *Bombus terrestris* pollination; degraded environments; growth cycle; *Lotus tenuis*; phytomanagement; seed multiplication.

The orchid flora of the Iron Gates Natural Park

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In Romania, the orchid family (Orchidaceae Juss., 1789) is represented by three subfamilies and 26 genera comprising 71 species. The Iron Gates Natural Park hosts a remarkable diversity of orchids. Traditional grasslands, preserved for centuries, cover around 10% of the park's surface and are of high conservation value due to their diverse plant associations. These habitats are a key component of the Danube Gorge landscape, though now fragmented and interspersed between forests and agricultural areas. This study aims to improve the knowledge of orchid distribution in the park, particularly for species that are nationally rare and of conservation priority. Fieldwork was conducted during the 2024 flowering season and early spring 2025. A total of 16 species and one intergeneric hybrid were identified. The most noteworthy findings include Orchis simia Lam., Orchis pallens L., Ophrys scolopax Cay. subsp. cornuta (Steven) E.G. Camus, Ophrys apifera Huds., Anacamptis papilionacea (L.) R.M. Bateman, Pridgeon & M.W. Chase, and Anacamptis × gennarii (Rchb.f.) H. Kretzschmar, Eccarius & H. Dietr., due to their rarity at the national level and their inclusion in the Red List of Vascular Plants of Romania. The temperate climate with sub-Mediterranean influences is reflected in the presence of taxa such as Anacamptis morio subsp. caucasiaca (K.Koch) H.Kretzschmar, Eccarius & H.Dietr., 2007, Anacamptis pyramidalis (L.) Rich., Limodorum abortivum (L.) Sw., Orchis mascula L. subsp. speciosa. (Mutel) Hegi. etc. The Iron Gates Natural Park remains a nationally important area for the conservation of orchid species restricted to this region. Further surveys are recommended to confirm the occurrence of *Himantoglossum calcaratum* (Beck) Schltr. and *Epipogium aphyllum* Sw.

Keywords: Conservation; Distribution; Iron Gates Natural Park; Orchidaceae; Romania

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Phytocoenological and population structure characteristics of Lysimachia maritima (L.) Galasso, Banfi & Soldano in Băile Someșeni (Cluj County, Romania)

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This paper provides insights into the population structure and ecology of the rare and endangered species Lysimachia maritima (L.) Galasso, Banfi & Soldano (syn. Glaux maritima L.) in the Băile Someseni area, a site with high ecological significance in Cluj County (Romania). This area is particularly important as it is the only known location in Romania where the species is clearly documented, featuring both halophytic and semi-halophytic plant communities. We investigated the population density and age structure of *Lysimachia maritima* in thirty plots across six transects following a moisture gradient. Additionally, stem height, number of flowers and fruits were recorded for reproductive individuals. To describe the vegetation, phytosociological relevés were carried out using the Central European phytosociological method. The following plant associations were identified: Scirpo-Phragmitetum W. Koch 1926, Triglochini maritimae-Asteretum pannonici (Soó 1927) Topa 1939, Plantagineto cornuti-Agrostetum stoloniferae Soó et Csűrös 1947, 1973, and Cladietum marisci Allorge 1921. Lysimachia maritima exhibited the highest density in phytocoenoses from Triglochini maritimae-Asteretum pannonici (119.75 individuals/m²), while plant communities from Scirpo-Phragmitetum showed the highest proportion of reproductive individuals (22.62%). Elevated values were also observed in transitional areas. Although reproductive individuals in Plantagineto cornuti-Agrostetum and Triglochini maritimae-Asteretum were taller and produced more flowers, fruit production and reproductive efficiency were the highest in *Scirpo-Phragmitetum* and in transitional areas. Based on our observations, the species appears to prefer moist soils. Litter cover significantly influences the population density, while the height of the herb layer affects the development of reproductive individuals and number of flowers. Given that this is the only known site in Romania where the species occurs, it was proposed in 2025 as a strictly protected area within the Natura 2000 network. Despite population stability, stronger conservation efforts are crucial, and further studies are needed to evaluate the impact of rising anthropic pressures on population dynamics.

Keywords: Băile Someșeni; halophytes; Lysimachia maritima; plant conservation; rare species

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Assessing climatic and topographic drivers of anatomical trait variation in *Carex curvula* across the European Alpine System

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Understanding how plants respond to environmental pressures is essential to predict their potential distribution. For this reason, in this study, we propose to evaluate the role of climatic and topographic drivers in determining anatomical traits variation in *Carex curvula*, a dominant sedge in primary alpine meadows across the European Alpine System. We used measurements of structural, vascular, and photosynthetic anatomical traits, and revealed that their variation is strongly related to environmental gradients. Under harsher conditions - such as high altitudes, increased aridity, and topographically exposed sites - Carex curvula exhibits a clear shift in allometry: leaves become smaller, with a higher investment in mechanical (sclerenchyma) and protective (epidermal) tissues, while the allocation to photosynthetic (parenchyma) tissues is reduced. This stress-adapted syndrome reflects a functional trade-off favouring mechanical reinforcement and water conservation over photosynthetic expansion, optimizing survival under environmental extremes. The observed shift aligns with the Leaf Economic Spectrum, emphasizing a strategy of resource conservation and tissue longevity rather than rapid growth. Altogether, our results underscore the important role of anatomical functional variability in enhancing the resilience of *Carex curvula*, providing crucial adjustments that would allow this species to persist under accelerated climatic changes along topographic gradients.

Keywords: alpine species, anatomical traits, Leaf Economic Spectrum

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Design and synthesis of novel ibuprofen-peptide conjugates for enhanced anti-inflammatory activity

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Ibuprofen (IBP), a non-steroidal anti-inflammatory drug is frequently administered for acute and chronic pain. However, the rapid absorption and short-halflife require frequent dosing. Despite the dose-dependently induced gastro-intestinal side effects, the oral route remains the most common and suitable route of delivery. Therefore, ongoing research aims to develop novel formulations to enhance the pharmacological profile of IBP. In the present study, novel conjugates of IBP with short peptides (IBP-L-Lys-L-Phe-L-Phe, IBP-L-Lys-D-Phe-D-Phe, IBP-L-Lys-L-Ala-L-Phe and IBP-L-Lys-D-Ala-D-Phe) have been developed using solid-phase peptide synthesis method and have been characterized through mass spectrometry (HPLC). The peptide content is expected to enhance the anti-inflammatory activity of IBP in vitro and to ensure antioxidant properties that could prevent gastro-intestinal mucosal lesions. Moreover, D isoforms of peptides could provide improved selectivity towards cyclooxygenases, target enzymes in the inflammatory pathway. Future investigations will focus on evaluation of cytotoxicity using the MTT assay, compared to the parent drug and free peptide sequences. Moreover, the pharmacological properties and the anti-inflammatory activity will be investigated using lipopolysaccharide-induced murine RAW 264.7 macrophages in comparison with the parent drug, by measuring ROS and pro-inflammatory cytokines production. Although further work is needed, the novel IBP-peptide conjugates could offer a promising strategy for improving pharmacological properties of IBP.

Keywords: drug design; ibuprofen; inflammation; peptides; solid-phase peptide synthesis;

Anticancer and antimicrobial bioactivity of extracts from black tulip, rosa, and peony: an *in vitro* assessment

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Natural plant extracts have gained significant attention as potential sources of bioactive compounds with therapeutic properties. This study investigated extracts from three ornamental flowers: black tulip (Tulipa sp.), rose (Rosa sp.), and peony (Paeonia sp.) for their anticancer and antimicrobial activities. The anticancer potential was evaluated through in vitro studies on melanoma and normal cell lines using WST-1 viability assays, LDH cytotoxicity measurements, ROS detection via confocal microscopy, and Western blot analysis of anti-inflammatory markers. Antimicrobial screening was conducted against nine reference bacterial strains, including Salmonella enterica (ATCC 14028), Shigella flexneri (ATCC 12022), Enterococcus faecalis (ATCC 51299), Escherichia coli (ATCC 25922), Pseudomonas aeruginosa (ATCC 27853), Staphylococcus aureus (ATCC 25923), Streptococcus pneumoniae (ATCC 49619), Streptococcus pyogenes (ATCC 19615), and two Candida species (ATCC 10231 and 22019). Results demonstrated differential cytotoxicity profiles, with black tulip extract exhibiting the most potent selective activity against melanoma cells while sparing normal cells. All extracts induced significant ROS generation in cancer cells and modulated inflammatory pathways. Antimicrobial assessment revealed broadspectrum activity, with rose extract showing the highest inhibition against gram-positive bacteria, particularly S. aureus and E. faecalis. This study highlights the potential of these ornamental flower extracts as promising sources of novel bioactive compounds with applications in both cancer therapeutics and antimicrobial treatments, warranting further investigation into their specific bioactive constituents and mechanisms of action.

Keywords: bacterial strains; cytotoxicity; melanoma; ornamental flowers; ROS.

Analysis of whole transcriptome data reveals the role of ELK3 in metastasis-associated processes in triple negative breast cancer

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Breast cancer is the most common cancer in women, with 2.3 million new cases yearly. 20% of which are triple-negative (TNBC). TNBC poses a major challenge due to its lack of key receptors, its highly invasive nature, and the absence of targeted therapies, making the development of such treatments crucial. This study aims to explore the role of the transcription factor ELK3 in cellular processes associated with metastasis in triple negative breast cancer cells, identifying the putative pathways modulated by ELK3. Genetic modification of MDA-231 cells was performed by lentiviral transduction to obtain cells with either ELK3 overexpression (ELK3-OE) or ELK3 knock-down (ELK3-KD). Genetic modifications were confirmed by fluorescence microscopy and flow cytometry via GFP+. Subsequently, a microarray analysis was performed in 4 biological replicas to obtain a whole transcriptome profile of the ELK3-KD cells. Both Ingenuity Pathway Analysis (IPA) and Gene Set Enrichment Analysis (GSEA) were used to identify putative pathways regulated by ELK3 in these cells. Lastly, 13 genes involved in cell motility and stemness were chosen to validate the microarray data by RT-qPCR. Over 95% of cells were successfully genetically modified in both ELK-OE and ELK-KD cell lines. Microarray analysis identified 740 genes modulated by ELK3. The functinal analyses based on molecular data predicted that cell migration and invasion were inhibited in ELK3-KD cells, with associated processes such as EMT and degradation of ECM being all supressed. Genes relevant for cell motility, namely ITGA6, STC1, VEGFA and LAMC2, were all found downregulated in ELK3-KD cells and overexpressed in ELK3-OE cells. In ELK3-KD cells, self-renewal capacity and key signaling pathways linked to cancer stemness (NFKB, JAK/STAT3, WNT/B-catenin, TGF-B, NOTCH1) were predicted to be inhibited. On the other hand, proliferation associated processes, including cell cycle progression, transition through the G2/M checkpoint and signalling through MYC and E2F were all activeted in cells with ELK3 supression. Genes involved in the acquisition of the stem-like phenotype (FGFR1, PTK2, WLS and WNT5A) and in cell proliferation (NDRG1, DUSP1, BNIP3L, MKI67 and SAE1) were modulated by both ELK-OE and ELK3-KD, indicating a potential regulatory relationship between ELK3 and these genes. According to whole transcriptome data, ELK3 enhances both the motility and the stem-like traits of TNBC cells, while hampering cell proliferation. These findings underscore the pro-metastatic role of ELK3 in TNBC.

Keywords: breast cancer; ELK3; metastasis: cell motility; cancer stem-like phenotype.

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SERS evaluation of anti-inflammatory activity of protein-based nonsteroidal anti-inflammatory drug (NSAID) conjugates

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Raman scattering spectroscopy is a well-known experimental technique due to its nondestructive nature, molecular specificity and ideal conditions for investigating biologic media for *in vitro* and *in vivo* studies. Herein were used NSAIDs, which are a class of drugs containing weak organic acids with similar chemical structures and inter-varying functional groups, as classical anti-inflammatory medication in comparison with newly designed drugs. Thus, to lessen the different side-effects like irritation or ulceration, ibuprofen (IB) has been conjugated with bovine serum albumin (BSA), by covalent binding, maintaining its parent drug antiinflammatory and analgesic activity. Because of Raman low sensitivity limitation, surfaceenhanced Raman scattering (SERS) technique was herein used for monitoring in real time the plasma samples collected from *in vivo* models subjected to chronic inflammation. The biomarkers specific to anti-inflammatory response were evaluated if present. The SERS spectral profiles of the samples collected from subjects treated with IB alone and IB-BSA conjugate. respectively. Results showed variations in terms of band shifting and intensity fluctuations for each scenario, providing useful insight regarding the novel NSAID action and side effects

Keywords: Chronic inflammation, ibuprofen, protein-drug conjugation. Raman spectroscopy, surface-enhanced scattering spectroscopy.

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Glycative Stress and Protein Functionality: New Aspects in Parkinson's Disease

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Parkinson's disease (PD) is a progressive neurodegenerative disorder marked by the loss of dopaminergic neurons in the *substantia nigra*. Although its precise mechanisms remain unclear, metabolic dysregulation and oxidative stress are recognised contributors. The pentose phosphate pathway (PPP), essential for generating ribose-5-phosphate and NADPH, supports glucose metabolism and cellular redox balance. However, its involvement in PD pathology is not fully understood. Particularly, the roles of ribose and xylose, two key PPP metabolites, have been insufficiently explored. This study investigates metabolic changes in PD with a focus on PPP intermediates. PD was induced in CD21 mice through intraperitoneal (i.p.) injections of 1methyl-4-phenyl-1,2,3,6-tetrahydropyridine (MPTP) at 25 mg/kg body weight. Experimental groups included MPTP-treated and control mice. After treatment, brains were dissected, and the cortical and striatal regions were isolated. Analyses included G6PDH activity, REDOX status markers, SDS-Page & PAS method, and metabolomic profiling of PPP-related metabolites, emphasising ribose and xylose levels. In MPTP-treated mice, significant alterations in oxidative stress markers and G6PDH activity were observed compared to controls. Metabolomic analysis revealed a statistically significant increase (p < 0.05) in ribose and xylose levels in the cortical and striatal regions. These results suggest that PPP dysregulation occurs alongside dopaminergic neuron loss in PD. study highlights a potential role for the pentose phosphate pathway in PD pathology. Elevated ribose and xylose concentrations in the PD model indicate that altered glucose metabolism via the PPP may contribute to oxidative stress and neuronal degeneration. These findings offer new insights into the metabolic underpinnings of neurodegeneration and suggest novel directions for targeted therapeutic strategies.

Keywords: glycation; metabolomics; neurodegeneration; Parkinson's Disease;

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Structural effects of P-glycoprotein missense mutations and pathogenicity of allelic variants

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P-glycoprotein (P-gp), also known as ABCB1 or MDR1, is an ATP-dependent efflux transporter involved in drug resistance and xenobiotic clearance. As a member of the ABC transporter family, it is primarily expressed in barrier tissues such as the blood-brain barrier, intestinal epithelium, liver and kidneys, where it limits intracellular accumulation of toxic substances. Structurally, Pgp includes two transmembrane domains (TMDs) forming a substrate-binding pocket and two nucleotide-binding domains (NBDs) that hydrolyze ATP to drive conformational changes for substrate transport. It has broad substrate specificity, transporting hydrophobic and amphipathic compounds, including drugs and peptides. Overexpression of P-gp contributes significantly to multidrug resistance (MDR) in cancer by decreasing the effectiveness of chemotherapy. In this study, 35 mutations with amino acid substitution were extracted from literature, tabulated, and analyzed for both structural impact and pathogenic potential. Pathogenicity was assessed using the AlphaMissense tool via the AlphaFold interface, while structural differences between native and mutant proteins were examined using Chimera, employing its matchmaker function for structural alignment. The analysis indicated no direct correlation between missense mutations and pathogenicity, suggesting that P-gp's disease relevance stems mainly from its role in drug resistance. However, several mutations caused noticeable local structural alterations. Further research is needed to clarify the link between these structural changes and alterations in protein stability.

Keywords: Missense mutations; pathogenicity; P-glycoprotein; structural changes

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Transcriptomic modulation in response to treatment with *in vitro*digested red cabbage-derived anthocyanins in colorectal cancer cells

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Anthocyanins, some of the most frequent active compounds in red- and purple-colored fruits and vegetables, are a class of dietary flavonoids with antitumor properties. They play a role in cancer prevention as well as in improving effectiveness of conventional therapies. The aim of this study was to examine the transcriptomic changes induced by *in vitro* digested red cabbagederived (Brassica oleracea var. rubra) anthocyanin extracts in DLD-1 colorectal adenocarcinoma cells by identifying affected cancer-related biological pathways. Control and IC50-treated DLD-1 cells were analyzed in four biological replicates. After 24h incubation with IC50 of the anthocyanin extracts, total RNA was isolated followed by transcriptomic profiling using onecolor gene-expression microarray slides. Differential gene expression analysis was conducted using the *limma* package in R. Gene set enrichment analysis (GSEA) was performed through clusterProfiler package using various gene sets (i.e. Hallmark, Reactome, KEGG). The objectives were : (1) to identify differentially expressed genes; (2) to assess the modulation of important cancer hallmarks, including proliferation, apoptosis, migration, and metabolism. A number of 10 genes involved in apoptosis and cell proliferation were randomly selected to validate the microarray data by RT-qPCR. A total of 420 genes were modulated by the treatment with the extract. The molecular data suggest that the anthocyanin metabolites from red cabbage supress cell proliferation and DNA repair, while activating pathways linked to apoptosis and p53 signaling. In addition, activation of the ROS pathway, glycolysis and hypoxia reflects profound metabolic stress. Apoptosis-related genes, namely CASP4, VHL, CXCL8 and SQSTM1 were overexpressed in anthocyanin-treated cells. Genes that are known to trigger cell proliferation (MYB, PDE4D, IFITM3 and BZW2) were inhibited, while CDKN1A and SDC4 genes that hamper cell proliferation were downregulated after treatment with the extract. In conclusion. anthocyanin metabolites from red cabbage seem to suppress multiple oncogenic pathways in colorectal cancer cells, showing promise in both cancer prevention and as complementary agents in cancer treatment.

Keywords: Anthocyanins; Colorectal Cancer; Transcriptomics

Paleopathological Finds from Medival Capidava

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Specimen C18 from the medieval burial site at Capidava (Topalu commune, Constanța county) displays several outstanding cranial and postcranial features. The pathological analysis of the cranial vault revealed osteolytic lesions, and medial meningeal vessel prints on the endocranium, a cranial deformation of the occipital and parietal bones congruent with plagiocephaly, as well as several supranumerary bones along the lambdoid and sagittal sutures. By analysing the occlusal plane of C18's molars, it was determined that the specimen had an open temporomandibular joint on the left-hand side as well as mandibular prognathism. The analysis of the postcranial skeleton displayed an incomplete fusion of the neural arches of L5 and sacral vertebrae, indicative of spina bifida occulta and an enchondroma present on a proximal phalanx, as well as noticeable wear of the cortical tissue of several bones. These features may indicate heritable growth and collagen defects, and several pathologies caused by collagen deficiencies were considered for a differential diagnosis: Ehlers-Danlos Syndrome, Osteogenesis Imperfecta, Marfan Syndrome, and Loeys-Dietz Syndrome.

Keywords: collagen defects; EDS; medieval; paleopathology.

Haemoprotein glycation: the Achilles heel of carbohydrate metabolism in neurodegeneration?

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A growing body of evidence suggests that there might be a stronger correlation between metabolic diseases and neurodegeneration¹. Insulin resistance and hyperglycaemia, characteristics in diabetes, could be pivotal in the instalment, development and progression of neurodegenerative pathologies. Shared molecular mechanisms such as glycosylation and glycation could also contribute to this link. Rat brain and hepatic mitochondria incubated with 40 mM sugar solutions (glucose, galactose, fructose and ribose) at 37 °C for 3 days showed distinct bands following SDS-PAGE and staining when compared to untreated mitochondria, implying the presence of glycated proteins. Moreover, Raman spectroscopy studies confirmed structural differences in proteins from treated versus untreated mitochondria. This primary observation led to subsequent experiments focusing on haemoproteins that could represent 'glycation' targets in glycating cellular stress conditions. Thus, we carried out studies on cytochrome c, monomeric haemoglobin (PDB accession numbers 5TY3, 1ECA) and compared these to human tetrameric glycated haemoglobin standards (Roche Diagnostics USA) used in clinical diagnostics. Protein solutions were incubated with the same cassette of sugars for 3 days at 37 °C. Samples were then analysed using UV-vis, SDS-PAGE and Raman spectroscopy, resulting in a plethora of evidence attesting the glycation of these proteins. Interestingly, galactose and ribose seem to be the sugars with the highest capacity for glycation out of the sugars studied. To extrapolate our results and make them relevant in the context of a broader range of brain related proteins (such as neuroglobin, cytoglobin etc.), bioinformatic analysis focusing on structural aspects is essential and will be conducted. We conclude that small proteins, specifically haemoproteins, are targets for glycation and modification in their structure could contribute to the instalment and development of metabolic related pathologies such as neurodegenerative disorders and hyperglycaemia. Furthermore, galactose has been observed to exhibit the highest binding affinity to the proteins studied.

Keywords: Glycation; Haemoproteins; Neurodegeneration

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Metal resistance strategies in terricolous micromycete, *Fusarium oxysporum* P2.7 isolate

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The spreading of heavy metals into almost any environmental compartment is inevitable with the ongoing technological development. Contamination by inorganic compounds is a serious hazard for human and ecological health. Remediation of heavy metals, unlike organic pollutants, is one of the most challenging tasks since these cannot be degraded and require complex interventions. Mycoremediation is an emerging biotechnology tool within bioremediation field, based on the properties of fungi to immobilize or reduce toxicity of heavy metals. Here, a highly metal-resistant Fusarium oxysporum isolate (GenBank ID: MT913528) was characterized for its resistance strategies against toxic concentrations (mg L^{-1}) of Cu (250), Hg (10), and Zn (150). The culture conditions were liquid medium, 28°C, darkness, active aeration for 7 days. The culture medium and biomass were acid digested and analysed by ICP-MS for metal content. Parallel biomass samples that were washed with a weak acid solution were employed to discern between the intra- and extracellular fractions. The micromycete's resistance against Cu and Zn stress was attributed to avoidance strategies, with limited to no intracellular uptake, since most quantity of these elements' interacting with the biomass was associated with the extracellular fraction. Moreover, the avoidance strategy for Cu was highlighted also on solid medium, where dendritic blue crystals formed at the edge of the growing colony. For Hg, extracellular immobilization was employed as a first defence strategy, sequestering Hg^{2+} in the cell wall, followed by permeation into the cytosol, where reduction to Hg⁰ followed by volatilization might have occurred. Understanding metal resistance mechanisms in fungi is a key step in developing mycoremediation as a valuable tool which can be tailored for a specific heavy metal. Further approaches should focus on both expanding understanding of metal resistance mechanisms by analysing transcriptome responses to metal stress and upscaling of the biomass application for remediation of relevant contaminated matrices.

Keywords: copper; Fusarium oxysporum; mercury; mycoremediation; zinc.

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