



The Balkan Botanical Congress is an international meeting that has been held nearly every three years, since 1997. It brings together botanists from around the world who perform research on plants in the widest sense, as well as scientists who are engaged in the plant sciences and their applications. We were honored to host such an extraordinary scientific event this year in Serbia.

The 7th Balkan Botanical Congress – 7BBC 2018 took place in Novi Sad from September 10th to 14th 2018. The Congress was organized by the University of Novi Sad, Faculty of Sciences, Department of Biology and Ecology and the “Andreas Wolny” Botanical Society, along with the great help of 7 co-organizers and more than 30 supporters and sponsors. It truly was not possible to happen without exceptional help of our co-organizer - the Institute for Nature Conservation of Vojvodina Province who made this congress not only possible, but totally awesome.

7BBC 2018 placed a special emphasis on plants of the Balkan Peninsula and covered various research fields. The Congress was organized into ten sessions: Plant Anatomy and Physiology, Plant Taxonomy and Systematics, Plant Molecular Biology and Genetics, Floristics, Vegetation and Phytogeography, Conservation Botany and Plant Invasions, Phytochemistry and Plant Resources, Agronomy and Forestry, Botanical Collections and History, Ethnobotany and Cryptogam Biology. These topics were elaborated through five plenary lectures given by eminent scientists, as well as in the form of introductory lectures, oral and poster presentations. With an overall number of 387 abstracts presented on the very latest of botanical science, we shared knowledge, expertise and novel ideas. We welcomed nearly 400 scientists to Novi Sad, and we believe that we succeeded in our joint endeavor to make new networks and new connections among botanists. We hope that we contributed to advancements in the wide and beautiful field of botany, ranging from fundamental botanical research to applied botany.

It is our great pleasure to publish this Abstract Book in Botanica Serbica, in the same year that this international journal, a renamed continuation of the Bulletin of the Institute of Botany and Botanical Garden Belgrade, celebrates its 90 year jubilee. On behalf of the Scientific and Organizing committee of 7BBC 2018 we would like to express our gratitude to all contributors, colleagues and sponsors for taking part in the 7th Balkan Botanical Congress, as well as for their efforts and contributions to it's successful realization.

Goran Anačkov and Lana Zorić,  
Co-presidents of the Scientific Committee of the 7 BBC  
and guest editors of Botanica Serbica 42 (supplement 1).

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### Sessions:

**The 7th Balkan Botanical Congress consists of plenary lectures, introductory lectures of each session, as well as oral and poster presentations on the following topics:**

**Sessions 1.** Plant Anatomy and Physiology

**Sessions 2.** Plant Taxonomy and Systematics

**Sessions 3.** Plant Molecular Biology and Genetics

**Sessions 4.** Floristics, Vegetation and Phytogeography

**Sessions 5.** Conservation Botany and Plant Invasion

**Sessions 6.** Phytochemistry and Plant Resources

**Sessions 7.** Agronomy and Forestry

**Sessions 8.** Botanical Collections and History

**Sessions 9.** Ethnobotany

**Sessions 10.** Cryptogam Biology

## Plenary lectures



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Plenary lecture 00 01

### TOWARD THE ANNOTATED CHECKLIST OF VASCULAR FLORA OF SERBIA – OBJECTIVES, METHODOLOGY AND CHALLENGES

Marjan Niketić<sup>1\*</sup>, Gordana Tomović<sup>2</sup> & Vladimir Stevanović<sup>3</sup>

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More than 30 years have passed since the first edition of the "Flora of SR Serbia" (1–10, 1970–1986) was published, continued with a new edition "Flora of Serbia" (1–2, 1992–2012). In the meantime, a significant number of papers have been published with new taxa for science and new floristic data for Serbia. Bearing in mind a certain number of taxonomic, chorological and floristic errors and inconsistencies in the mentioned editions, as well as a new extensive floristic material collected in the last 30 years, there was a need for the "Annotated Checklist of Vascular Flora of Serbia", which would include all new contributions and recent nomenclatural, taxonomic, chorological, phylogenetic and phylogeographic points of view on plant taxa. The most informative part of the Checklist would be a catalog with cited chorological and taxonomic-nomenclatural sources, including a dozen of relevant international plant lists and other publications. Considering that there are a significant number of different concepts in them, it has been found that as many as 34% of the plants still have an unresolved taxonomical status. For processing data from database, the following software applications were created: correction, navigation, searching, data crossing, status determination and sorting. Applications for output reports can also be divided into six categories: catalogue, synonyms, comments, form for authors, literature and statistics form. Seven literary and herbarium databases with over 570,000 records were used to determine the presence of taxa by certain territorial units (Central Serbia, Vojvodina, Kosovo and Metohija). Preliminary statistics show that the presence of 4246 taxa (species, subspecies and hybrids) was confirmed for the territory of Serbia, of which 3690 species. Of these taxa, 192 (4.5%) are allochthonous, with exclusion of plants escaped from cultivations, considered as ephemeral aliens that still not established self-reproduced stable populations. There are 200 plant taxa described from Serbia, of which 51 species and subspecies are considered national endemics. For an additional 438 taxa the presence in the Serbian flora is disputed, 44 taxa are considered to have disappeared, while 17.2% of the taxa was not mentioned in two editions of the „Flora of Serbia“.

**KEYWORDS:** vascular flora, Checklist, Serbia

Plenary lecture 00 02

### TYPES OF ORGAN FUSION IN ANGIOSPERM FLOWERS (WITH EXAMPLES FROM CHLORANTHACEAE, ARALIACEAE AND MONOCOTS)

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Fusion between floral organs or their parts is believed to have played key roles in origin and subsequent diversification of angiosperms. Two types of fusion are recognized: postgenital and congenital. Postgenital fusion is really observable during flower development: primary morphological surfaces of contacting structures meet and join during this process. After *perfect* postgenital fusion, no trace of the original epidermal layers can be recognized, but these remain visible, often in modified form, after *imperfect* postgenital fusion. Congenital fusion cannot be directly observed and takes place due to differential growth. In the case of *complete* congenital fusion, free parts of fused organs cannot be seen at any developmental stages. *Incomplete* congenital fusion implies presence of free organ parts on the common (united) base; it can be divided into *early* and *late* congenital fusion depending on whether the common base precedes or follows the initiation of free parts during development. Phenomena related to congenital fusion are development of free organs from common primordia, hybridization of developmental pathways, loss of organ individuality, heterotopies and fasciation. Differences between congenital and postgenital fusion are much more unequivocal than those between the presence and the absence of fusion. There is no abrupt boundary between imperfect postgenital fusion and transient contact between organs during development. Structures assumed to be congenitally fused clearly develop as a unit, but it is necessary to demonstrate that these structures indeed belong to different merged organs (instead of being parts of the same organ or two distinct organs on a common base). This only can be done in the framework of comparative morphology. Analyses of both types of fusion involve arbitrary decisions, so it is not appropriate to discard existence of any type. Conventional interpretations of morphological concepts lie at the base of analyses of character evolution, even if they are performed using maximum parsimony or model based methods and molecular phylogenetic data. Patterns of organ fusion are discussed here using three case studies.

**KEYWORDS:** androecium, angiosperms, Apiales, Araliaceae, calyx, Chloranthaceae, congenital fusion, corolla, gynoecium, *Hedyosmum*, monocots, postgenital fusion, septal nectarines

Plenary lecture 00 03

### POLYPLOID COMPLEXES IN THE (SUB-)MEDITERRANEAN: EXAMPLES FROM THE GENUS *CARDAMINE*

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Polyploidisation, a process of rapid evolution through genome duplication, has played a key role in evolution and diversification of living systems and flowering plants in particular (all extant angiosperm lineages experienced at least one episode of genome duplication). It was suggested that polyploidisation even may be the most common mechanism of sympatric speciation in plants. For many polyploids, multiple origins in space and time have been shown, increasing their genetic diversity and complexity, whereas in other cases a single origin has been proposed. Although the wealth of knowledge about plant polyploidy has increased significantly in last decades, there are considerable gaps in our understanding of the general evolutionary triggers and consequences of polyploidisation in natural populations. Overcoming the gap between artificial models and natural systems is crucial for our understanding of polyploid evolution from a complete perspective, including the genuinely practical outcomes (e.g., development of artificial polyploid crops capable to thrive in natural conditions). The Brassicaceae family may provide the most suitable study systems – while the patterns in few model complexes are comparatively well-known (e.g., *Arabidopsis*, *Brassica*), the investigations of their wild relatives are just ripe to get our sufficient attention. Here I present several examples of studies of polyploid complexes in the polyploidy-rich genus *Cardamine* of the Brassicaceae family from the (Sub-)Mediterranean area. Indeed, the area of the Mediterranean and its surroundings played an important role in the evolution of numerous polyploids and polyploid complexes and the genus *Cardamine* is not an exception here. Studies presented here were based on results of the chromosome counting, flow cytometry, multivariate morphometrics, Sanger sequencing, microsatellite and fragment analyses and, most recently, also of the next generation sequencing. Although some cases of autopolyploids were found (e.g. *C. majovskyi*), in most cases complex polyploid origin was confirmed (e.g., *C. asarifolia*). While in the Mediterranean and Sub-Mediterranean the genus is represented mostly (but far not exclusively) by diploids, north of this area mostly polyploids occur, up to the dodecaploid level. Apart from regular eupolyploidy, dysploid and aneuploid plants were found. Hybrid individuals were proven as well as triploids resulting from fertilization by unreduced gametes.

**KEYWORDS:** Brassicaceae, *Cardamine*, Cruciferae, glacial refugia, polyploid complexes

Plenary lecture 00 04

### GRASSLAND RESEARCH AND CONSERVATION WITHIN THE EURASIAN DRY GRASSLAND GROUP (EDGG)

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Grassland ecosystems are home to biodiversity, sources of natural beauty and cultural values. Due to recent land-use changes and gradual vanishing of traditional rural cultures, eutrophication from agricultural fertilization, but also airborne nitrogen deposition from multiple sources a rapid loss of biodiversity and homogenization are detectable in almost all types of Palearctic grasslands. Eurasian Dry Grassland Group (EDGG) is an international organization dealing with research and conservation of Palearctic grasslands, with more than 1200 members from nearly 70 countries. During 2008-2017 I served as one of its chairs and was included in numerous of its activities. In my talk I will focus on those with major contribution to grassland research and conservation. Grassland research was supported mainly by preparation of special issues on grassland-related topics in international scientific journals, organizing research fieldworks and expeditions, and establishment of the GrassPlot database. Grassland conservation initiatives such the Smolenice Grassland Declaration highlight the necessity of a strong and comprehensive international frameworks and well-developed national strategies for grassland conservation, which are still missing.

**KEYWORDS:** biodiversity, database, GrassPlot, Palearctic, research expedition, special issue

Plenary lecture 00 05

### TRENDS AND CONSEQUENCES OF BIOLOGICAL INVASIONS IN THE ANTHROPOCENE

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Biological invasions have become a defining feature of global environmental change. However, the global patterns and underlying factors that determine variation in invasions worldwide are still insufficiently understood. Similarly, the consequences and future trajectories of biological invasions are not fully appreciated. However, in the last years, the substantial increase in global distribution data on alien species of several taxonomic groups has for the first time allowed to compile databases which provide accurate and exhaustive information on the global richness, flows between regions, taxonomic composition and temporal accumulation of alien species. Similarly, the compilation of the Alien Species First Record-database provides a backbone for analysing spatio-temporal patterns of alien species accumulation. Further, data on anthropogenic pressures, on the exchange routes of goods and people, and on a large range of environmental factors have increasingly become available. Combined, these data sets allow for robust analyses of the patterns and processes of invasions world-wide and they provide the foundation for exploring future trends of alien species spread and the impacts these may cause. In this talk, I will present key insights that have emerged from these novel global databases. Further, I will highlight likely future consequences of biological invasions, and identify gaps in knowledge which have to be addressed as a priority.

**KEYWORDS:** alien species, anthropogenic pressures, Alien Species First Record-database

## Session 1. Plant Anatomy and Physiology



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Introductory lecture 01 01 07  
**MORPHOGENETIC EFFECT OF GENOME SIZE  
IN TRIBE FABEAE (FABACEAE)**

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Evolution of living things is thought to be driven mostly by changes in genes and resulting phenotypic novelties subjected to natural selection. However, except for gene mutations, a genome may undergo other changes, e.g. amplification of non-coding regions, such as repeats or transposable elements. Such events lead to increase in genome size (C-value), which, in its turn, has some effects on cell properties regardless of the information content of a nucleus. These effects are termed “nucleotypic”. Among obvious consequences of increase in nuclear DNA content, one may expect the enlargement of cells and extension of cell cycle due to longer replication time. A given work aims at detection of “nucleotypic” effects in tribe Fabeae (Leguminosae). This group includes five genera, two species-rich (*Lathyrus*, *Vicia*) and three species-poor (*Lens*, *Pisum*, and *Vavilovia*). These plants are exceptional as having the largest C-values among Leguminosae, although polyploidy is rare in this tribe. They inhabit subtropical and temperate regions and many species of Fabeae are of significant practical value, such as pea, lentil, broad bean etc. C-values were extracted from the Plant DNA C-values Database, while other parameters were obtained in course of presented work. C-value correlates with sizes of pollen grain, stomata, epidermal cells, flowers, seeds, stem apical meristems, and thickness of seed coat. These correlations were of different reliability (and sometimes even oppositely directed) in *Lathyrus* and *Vicia*. Moreover, meanings of C-value are distributed non-randomly between annual and perennial species of Fabeae. Amplification of non-coding elements of genome could be one of the main reason for specificity of this taxon, possibly even affecting its geographical distribution.

**ACKNOWLEDGMENTS:** The work was partially supported by the Russian Foundation for Basic Research (project no. 18-34-00511).

**KEYWORDS:** C-value, *Vicia*, *Lathyrus*, evolution, ontogeny

Introductory lecture 02 01 51  
**INFLUENCE OF PHOSPHORUS FERTILIZA-  
TION ON ARBUSCULAR MYCORRHIZAL COL-  
ONIZATION AND PHOSPHATASE ACTIVITY  
ON LENTIL (*LENS CULINARIS* MEDIK.)**

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Due to the low soil availability of phosphorus (P), various mechanisms are employed by plants, including vesicular arbuscular mycorrhizal (VAM) fungi and phosphatase enzymes, in order to increase its uptake. The aim of this work was to study the effect of P fertilization on VAM colonization, phosphatase activity and seed yield of lentil. A field experiment was conducted for two growing seasons (Gs) (2014 and 2015) in the farm of Aristotle University of Thessaloniki. Under rainfed conditions, four lentil cultivars were pre-seeding supplemented with four P rates (0, 30, 60 and 90 kg P<sub>2</sub>O<sub>5</sub>/ha) in a split-plot design with P rates in the main plots and cultivars in the subplots and three replications. Samplings of above-ground biomass, roots and the rhizosphere took place at six growth stages, two vegetative and four reproductive. Dry weight, colonization of VAM, alkaline and secreted acid phosphatase activity was determined for each subplot. At full maturity, seed yield was determined. Both VAM colonization and seed yield were significantly affected by P rates, cultivars, Gs and the interaction cultivar×Gs. The highest VAM percentage was recorded in 2015 (40%) when seed yield was almost double (4345.6 kg ha<sup>-1</sup>) compared to 2014. For the cultivar×Gs interaction a positive correlation between VAM colonization and seed yield was found (r=0.83, n= 8, P<0.01). VAM colonization increased during the vegetative stages and reached a maximum at full bloom (44.1%), with a significant decrease thereafter. Unlike VAM, the activity of both acid and alkaline phosphatases was gradually decreased with the progress of Gs (from 192.9 to 113.3 PN μg g<sup>-1</sup> h<sup>-1</sup> and from 289.9 to 256.6 PN μg g<sup>-1</sup> h<sup>-1</sup>, respectively). Phosphorus rates had no significant effect on phosphatases but they were significantly affected by the interaction cultivar×Gs. For the cultivar×Gs interaction, a strong negative correlation between VAM colonization and acid phosphatates was evident (r= -0.93, n = 8, P< 0.001). In conclusion, P had no effect on rhizosphere activity, but VAM colonization seemed to affect root phosphatases.

**KEYWORDS:** growth stages, rhizosphere activity, yield

Oral presentation 03 01 16

### THE VARIABILITY OF LEAF SILICON AND CALCIUM CONTENT IN *DESCHAMPSIA CESPITOSA*

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Silicon (Si) and calcium (Ca) are regarded as important plant biominerals, as they affect plant water and energy balance, and improve plant resistance to pathogens and herbivores. Grasses exhibit high contents of some biominerals such as Si in their tissues. Biominerals mainly accumulate in the epidermis and thus significantly affect leaf optical properties. Their uptake is highly dependent on transpiration flow. *Deschampsia cespitosa* L. is a perennial grass that is found in many grassland types, but favours poorly-drained soils. The objectives of this study were to examine various leaf traits in *D. cespitosa* from different locations, especially the contents of Si and Ca in the leaves and the corresponding soils. Plants were sampled from four different locations, namely floodplain of the river Rak near Rakov Škocjan and intermittent Lake Cerknica, these two being characterised as wetland sites on carbonate rocks. In contrast, two sites with prevailing calc-alkaline volcanic rocks were chosen, namely a heath under the top of the mountain Komen and a forest edge at the foothills of the same mountain. Ten plant and soil samples were collected from each location. Leaf morphological properties were analysed along with their reflectance and transmittance. Moreover, chlorophyll a, chlorophyll b, carotenoid, anthocyanin, and UV-absorbing compounds leaf contents were determined. The concentrations of Si, phosphorus (P), sulphur (S), chlorine (Cl), potassium (K), and Ca in the leaves and soils were determined using X-ray fluorescence spectrometry. The soil properties differed significantly between the four locations, including their element composition. Contrasting habitat characteristics resulted in differences in leaf morphological, biochemical, and optical properties of samples from different locations, along with their leaf element contents. These differences were related to rock and soil type. Plants from the heath differed substantially from plants sampled from other locations. This study revealed that differences in habitat characteristics significantly affect plant leaf traits, including their amount of biominerals. Considering the significance of Si for grasses, this could affect their fitness in specific environments.

**KEYWORDS:** tufted hairgrass, rock type, soil properties, biominerals, element analysis

Oral presentation 04 01 28

### THE RESPONSE OF KOHLRABI SPROUTS (*BRASSICA OLERACEA* L. VAR. *GONGYLODES* L.) TO THE ADDITION OF SELENATE AND SELENITE

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Selenium (Se) is deemed as an essential element for humans and animals, however, its essential role for plants has not yet been established. It is known that Se has a positive effect on plants in lower concentrations, while it can have a negative impact in higher concentrations. Slovenia is a country with low concentrations of Se in the soil. The aim of the present research was to determine the effect of different forms of Se on selected biochemical and morphological characteristics of kohlrabi sprouts (*Brassica oleracea* L. var. *gongyloides* L.). Sprouts were grown from seeds which were soaked in solutions of selenite and selenate. Seeds germinated in a growth chamber in controlled conditions. At the end of the experiment, we measured the content of chlorophyll a and b, carotenoids, anthocyanins, and UV-A- and UV-B-absorbing compounds. Photochemical efficiency of photosystem (PS) II was measured as well. In order to compare the results, this experiment was carried out twice. Sprouts treated with selenate synthesized lower amounts of chlorophyll a and carotenoids compared to sprouts that germinated from seeds which had been soaked in selenite. The chemical form of added Se did not affect synthesis of UV-B- and UV-A-absorbing compounds. Potential photochemical efficiency of photosystem II was close to the theoretical maximum (0.83) in both control and treated sprout groups. Selenate treatment had a positive impact on the dry weight of sprouts. None of the selenium treatments presented stress conditions for experimental sprouts.

**KEYWORDS:** kohlrabi, sprouts, selenate, selenite

Oral presentation 05 01 02

### NITRIC OXIDE ALLEVIATES CADMIUM TOXICITY ON BLACK POPLAR (*POPULUS NIGRA*): PHYSIOLOGICAL APPROACHES

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Heavy metals are dangerous environmental pollutants and their toxicity causes health hazards for plants survives. Plants are exposed to heavy metal pollution in the environment. Cadmium (Cd) is non-redox toxic heavy metal found in soil, water, and air. Cd is taken up from soil by plant roots and transported into leaves by xylem loading, resulting in toxicity symptoms, such as growth reduction, wilting, chlorosis, and cell death. Nitric oxide (NO) is a gaseous signaling molecule in plants that plays important roles in wide range of molecular and physiological processes in responses to biotic and abiotic stresses. In present study, we have investigated the effects of exogenous nitric oxide (NO) supplementation as sodium nitroprusside (SNP), on Cd toxicity in black poplar (*Populus nigra*) and thus protective role against Cd toxicity. The black poplar was exposed to Cd stress individually (100 and 500 µM CdCl<sub>2</sub>) or in combination with SNP (100 µM SNP) for a period of 28 days by using complete randomized design with three replications. The all treatments with Cd exposure significantly reduced chlorophyll contents (Chl a, Chl b, Chl a+b), but NO treatments increased total chlorophyll contents. The addition of NO also reduced the malondialdehyde (MDA) and hydrogen peroxide (H<sub>2</sub>O<sub>2</sub>). Cd concentrations decreased in root and bark of *P. nigra* with NO treatments, whereas Cd leaf concentrations increased in relation with supplied NO. The NO supplied by the SNP enhanced the dry weights of roots, leaves, and bark. While the bioconcentration factor (BCF) decreased in the root and bark, an increase in the leaves was detected with NO treatments. It was also found that Cd translocation factor (TF) in leaves increased by NO application and decreased by NO treatment in bark. The study revealed that an exogenous supply of NO protects black poplar from Cd-induced toxicity and improves some physiological parameters.

**KEY WORDS:** Cadmium toxicity, sodium nitroprusside, photosynthetic pigments, stress physiology

Oral presentation 06 01 52

### EVALUATION OF MAIZE INBRED LINES AND HYBRIDS UNDER DROUGHT STRESS USING PHYSIOLOGICAL TRAITS

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Breeding for maize drought tolerance has always been a significant objective for breeders and plant physiologists. Nowadays, climate change sets new challenges to major crop adaptation at stressful environments. For such a purpose, the measurement of physiological traits related to maize response to drought might prove to be useful indices. The objective of the present study was to establish whether the physiological traits can be used as reliable physiological markers to evaluate the performance of parental genotypes and their hybrids under both dry and normally watered conditions, at different growth stages. Four (4) maize inbred lines and 5 single-crosses among them were evaluated under normal and water deficit regime (50%), across two locations at northern Greece for two growing seasons (2016 and 2017). Measurements [leaf chlorophyll content (SPAD) and photosynthetic parameters] were taken at specific growth stages and at full maturity grain yield was calculated. Treatment with limited irrigation had a negative effect to all physiological traits and caused a 40% yield lost SPAD values had a significant positive correlation with the grain yield only for the reproductive stages, with the highest correlation to be found at the R2 stage ( $r=0.61$ ,  $n=60$ ,  $P<0.01$ ). The photosynthetic measurements showed significant positive correlation with grain yield at the R2 stage ( $r=0.66$ ,  $n=60$ ,  $P<0.01$ ). On contrary no statistical correlation observed at vegetative stage for both SPAD and photosynthetic measurements. In conclusion, physiological characteristics can facilitate the selection of stress-adaptive genotypes especially when they are used during the reproductive stages.

**KEYWORDS:** photosynthesis, SPAD, water use efficiency

Poster presentation 07 01 29

**MICROMORPHOLOGICAL AND ANATOMICAL VARIABILITY OF *ASTRAGALUS MONSPESSULANUS* L. AND *A. SPRUNERI* BOISS. (FABACEAE) FROM THE CENTRAL PART OF THE BALKAN PENINSULA**

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The genus *Astragalus* L. is one of the largest genera of vascular plants which, besides numerous researches still represents a genus with not enough explored taxonomic problematic. Many species are narrowly endemic, while relatively small number of them are wide-spread, distributed in the northern hemisphere, especially in Central Asia and Western North America. In the present study, two similar species, *A. monspessulanus* and *A. spruneri*, which belong to section *Incani* DC. from the area of central Balkans were studied. Due to the high polymorphism the section is taxonomically the most problematic group of the genus and the boundaries between its representatives are still not completely clear. The aim of this study was to determine the micromorphological and anatomical variability of populations of these species based on quantitative characters of leaves and petioles, and also, to show to the potential degree of differentiation within species. Characters were analyzed on individuals from 6 populations (4 populations of *A. monspessulanus* and 2 populations of *A. spruneri*). Statistical analyses were carried out for 32 quantitative characters related to the leaf and petiole anatomy. Data analysis included the application of analysis of variance (ANOVA), discriminant analysis (DA) and cluster analysis (CA). The results indicate a high variability of analysed characters and anatomical differentiation of populations of the species, and at the same time, the clear separation among studied species.

KEYWORDS: *A. monspessulanus* L., *A. spruneri* Boiss., anatomical characters, differentiation

Poster presentation 08 01 19

**MORPHOLOGY AND ANATOMY OF AN ENDEMIC *ALCHEMILLA ORDUENSIS* PAWL. (ROSACEAE) SPECIES FROM TURKEY**

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A critical and taxonomically difficult group, the genus *Alchemilla* L. of the family Rosaceae includes more than 1000 species with a holarctic distribution in the world. In Turkey, the genus is represented by 73 species. The taxonomical research on this genus has continued on a large scale to present day. However, despite extensive taxonomical studies, anatomical data concerning *Alchemilla* are still scarce. In the present study, an endemic *A. orduensis* Pawl. species distributed in north-east Anatolia (Turkey) was investigated in point of morphological and anatomical features. In the morphological part of study, the external morphology of the leaf and flower was determined from herbarium materials deposited at KTUB (Karadeniz Technical University Biology Department). In the anatomical studies, the anatomy was examined on transversal sections of the stem and leaf. The stem anatomy of *A. orduensis* consists of single layered epidermis, 2-3 layered collenchyma, a sclerenchyma ring placed between endodermis and phloem. The pith comprises large orbicular or polyhedral parenchymatic cells. The leaf anatomy of the species is determined as lower- and upper epidermis, palisade parenchyma and spongy parenchyma. Stomata are of anomocytic type and vascular bundles are of collateral type, surrounded by a parenchymatic bundle sheath. The anatomical characters of this endemic species is reported for the first time. The data are discussed in terms of taxonomical point of view.

KEYWORDS: Rosaceae, *Alchemilla*, morphology, anatomy, Turkey

Poster presentation 09 01 49

**ANATOMICAL VARIABILITY OF POPULATIONS OF *FESTUCA VALESII* COMPLEX (POACEAE) IN WESTERN AND CENTRAL SERBIA**

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*Festuca valesiaca* complex represents Eurasian group of species (*F. callieri*, *F. dalmatica*, *F. panciana*, *F. rupicola*, *F. stajanovii*, *F. stricta*, *F. trachyphylla*, *F. valesiaca*, *F. wagneri*, etc.), with center of distribution and diversity in the steppe regions of Asia and Eastern Europe. The group is taxonomically intricate, and it is hard to differentiate taxa based only on the

characters of the outside morphology of vegetative and reproductive organs. However, the anatomical characters of the leaves, as in many other grasses, can help in distinguishing the closely related species. So far, the research on distribution, as well as the morpho-anatomical characteristics of populations of *F. valesiaca* complex on the Balkan Peninsula has not been conducted. Therefore, the aim of this study was to describe the variability of the characters of the leaf anatomy in different populations of *F. valesiaca* complex on the territory of Western and Central Serbia. Morphometric analysis was performed on cross-section of 191 tiller leaves collected from 13 populations. Descriptive statistics (mean, standard deviation, minimum, maximum and standard error, coefficient of variation) was calculated in detail for 14 anatomical characters. Analysis of variance (ANOVA) was performed to identify significant variation between each character. Principal component analysis (PCA) based on the complete dataset was done to show the overall morphological variation. Finally, canonical discriminant analysis (CDA) using populations as a priori groups, and cluster analysis based on Mahalanobis distances were performed as well. The results of our study showed that the highest number of analyzed anatomical characters have a moderate degree of variation, with coefficient of variation between 20 and 50%. Analysis of variance showed that all the characters have statistically significant contribution to the differentiation of analyzed populations. Principal component analysis has shown that individuals in analyzed populations constitute a homogeneous group, suggesting complex anatomical variability of the studied populations. Although discriminant analysis showed significant overlapping of the populations, cluster analysis identified three groups that could be assigned to independent taxa. Further more detailed morphometric and molecular analyses are needed in order to examine the complexity of this intricate Eurasian group of fescues.

KEYWORDS: *Festuca valesiaca*, leaf anatomy, Poaceae, Serbia, variability

Poster presentation 10 01 35

**STUDY ON *GALANTHUS* SPECIES IN THE BULGARIAN FLORA**

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Genus *Galanthus* (Amaryllidaceae) includes 19 species naturally distributed in Europe and Middle East. The Flora of Bulgaria recognizes two species: *G. nivalis* L. and *G. elwesii* Hook. *Galanthus elwesii* is characterized with relatively high

morphological variability, leading some authors to identify some populations as *G. gracilis* Celak. However, there are no research reports confirming the occurrence of *G. gracilis* in Bulgarian flora. Therefore, the objective of this study was to compare *G. nivalis* and *G. elwesii* with plants from populations previously identified as *G. gracilis*. For this purpose, morphological, DNA, embryological and anatomical studies were carried out. The morphological characteristics and DNA dendrogram revealed that *G. gracilis* and *G. elwesii* were situated in the same cluster and had significant morphological similarity, whereas plants from populations identified as *G. nivalis* were dissimilar in morphology and situated in a separate cluster. The revealed features of the generative sphere showed similarities across the species, and namely: tetrasporangiate anthers, Monocotyledonous-type of development of the anther wall, two-celled mature pollen grains; Polygonum-type of development of the female gametophyte; Asterad-type embryogenesis. Anatomical studies of the leaf structure of the three species, revealed a single-layer epidermis, stomata situated on the abaxial and adaxial surface of the leaves, a similar mesophyll with lysigenous cavities of various sizes. The anticlinal walls of *G. elwesii* and *G. gracilis* were straight, while these of *G. nivalis* were wavy. This research demonstrated that the plants of *G. elwesii* and plants from populations identified as *G. gracilis* are morphologically, embryologically and genetically similar. This study did not provide sufficient evidence to support the claim on the existence of *G. gracilis* in the Bulgarian flora; the populations identified as *G. gracilis* in Bulgaria may be forms of *G. elwesii*.

ACKNOWLEDGEMENTS: The present study was supported by the Scientific Research Centre of the Agricultural University Plovdiv: Project №12-15 awarded to Dr. Semerdjieva and colleagues.

KEYWORDS: *Galanthus*, morphology, DNA, embryology, anatomy

Poster presentation 11 01 32

**ANATOMICAL INVESTIGATIONS OF *ARTEMISIA CAMPESTRIS* L. (COMPOSITAE) FROM MONTENEGRO**

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Genus *Artemisia* L., the largest genus of the tribe Anthemideae (Compositae), comprises more than 500 taxa at specific or subspecific levels. Infrageneric taxonomy of *Artemisia* genus is very complex because some species have very differ-



ent morphological forms while others closely resemble each other. In this work, anatomical investigations of vegetative organs of *Artemisia campestris* L. (Compositae), wild-growing species from Montenegro, were conducted. The aim of this study was to examine the general anatomy and particular anatomical features which are in relation with production of specialized metabolites, as well as to find possible new valid taxonomic characters. Microscopic slides were prepared following the standard histological procedures. Typical secondary growth occurs in the older root with small group of sclerenchyma fibers scattered in the cortex. The stem cross section is characterized by more or less round shape with collateral vascular bundles. The largest parenchyma cells, with lignified cell walls, occur in the pith. Below the epidermis, well developed hypodermis could be noticed, made up of a several layers of enlarged cells arranged in radial rows. Leaf cross section has triangular shape, with well developed cuticle and the isolateral palisade structure. Secretory ducts are present in the root and stem cortical parenchyma, as well as in the leaf parenchyma. The stem and leaves are almost glabrous, with a very few trichomes that are noticed. All of the obtained data, may be considered as possible valid taxonomic characters which could be helpful in species identification and infrageneric taxonomy of the genus *Artemisia*. Thus, these findings are of importance for future anatomical, micromorphological and phytochemical investigations of this and related species of the genus.

**KEYWORDS:** *Artemisia campestris*, Asteraceae, anatomy

Poster presentation 12 01 37

### MORPHO-ANATOMICAL DISTINCTION OF SIMILAR TAXA: *INULA BRITANNICA* L. AND *I. OCULUS-CHRISTI* L.

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*Inula britannica* and *I. oculus-christi* are well defined species, but morphological similarity between them is notable and, indisputably, they resemble to each other. Due to their morphological variability conditioned by environmental factors, specimens of each have commonly been misidentified. In the present study, detailed morpho-anatomical analyses of leaf, stem, rhizome, fruit and receptacle of *I. britannica* and *I. oculus-christi* were performed. The objective was to define further characters that would be useful in taxonomic delimitation of those species. Plant material was collected from native populations and each of the species was collected from the five localities, ten samples from ten different individuals belonging to each population were analysed. Cross-sections

of leaves, stems, rhizomes and fruits were obtained using cryotechnique procedure by Leica CM 1850 cryostat. Detailed descriptions of anatomical characteristics of the analyzed organs were given using light microscopy, while Scanning Electron Microscopy revealed specific qualitative features of leaf, fruit and receptacle that characterized each species. Our results indicated the taxonomic relevance of the leaf and fruit indumentum, presence of trichome types on receptacle surface, fruit and pappus dimension, number of fruit sclerenchymatous bundles, organization of stem chlorenchyma and number of vascular bundles on stem cross section, in distinguishing these two species. Obtained data are useful in species identification and represent the valuable contribution to morpho-anatomical differentiation of those congeneric, commonly misidentified, taxa.

**KEYWORDS:** anatomy, micromorphology, *Inula*

Poster presentation 13 01 18

### DEVELOPMENT, ANATOMY AND HISTOCHEMISTRY OF LEAF TRICHOMES OF *INULA BRITANNICA* L. (ASTERACEAE)

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*Inula britannica* L. (Asteraceae) is an erect biennial or perennial plant distributed in Europe and Asia, where it typically thrives in a variety of moist habitats. The genus *Inula* is known for its great biological activity, which is based on a large number of secondary metabolites that plants synthesize and accumulate mostly in glandular trichomes. Accordingly, the aim of present study was to describe the development, structure and chemical composition of the secreted material of leaf glandular trichomes of *in vitro*-grown *Inula britannica*. Morpho-anatomical and histochemical analyses of the leaf trichomes were carried out using conventional light and Raman spectroscopy. On both leaf surfaces of *in vitro* *Inula britannica* plants, two classes of trichomes were recognized: mechanical and glandular capitate trichomes. Glandular trichomes started as outgrowths of epidermal cells; first periclinal division generated the upper and lower cells. Subsequent divisions of the upper cell gave rise to stalk and secretory cells of glandular trichomes. The mature glandular trichomes were multicellular and biseriate with two epidermic basal cells, 8–10 penduncle cells and secretory head composed of two cells. Histochemical analysis has shown that different substances were synthesized and accumulated in the leaf glandular trichomes of *in vitro* *Inula britannica*. Lipids, terpenes and alkaloids were stored in a large subcuticular space of glandular trichomes. Besides,

the Raman spectroscopy performed directly on the tissues (in situ) confirmed the presence of terpenes, probably the bicyclic monoterpenes ( $\alpha$ -pinenes, recognized according to the highest intensity band at 1653 cm<sup>-1</sup>) and sesquiterpenes, in the subcuticular space of the leaf trichomes, whose presence indicated the strong intensity band at 1434 cm<sup>-1</sup>.

**ACKNOWLEDGMENTS:** This work was supported by projects 173015, 173024 and 31005 of the Ministry of Education, Science and Technological Development of the Republic of Serbia

**KEYWORDS:** *Inula britannica*, glandular trichomes, Raman spectroscopy, aromatic plant, secondary metabolites

Poster presentation 14 01 33

### PHYLLARY CHARACTERISTICS OF WILD PERENNIAL *HELIANTHUS* L. SPECIES

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Phyllaries or involucre bracts surround capitulum of *Helianthus* L. species, play an important role in dispersion, pollination, seed germination and may protect the inflorescences against predators and negative effects of environmental factors. The aim of this research was to conduct comparative micromorphological and anatomical analyses of phyllaries of 19 wild perennial *Helianthus* species. Plant material was hand-collected at the time of physiological maturity. For anatomical analyses cross-sections were obtained from the middle part of the phyllaries using cryotechnique procedure. Micromorphological observation, morphological and histological measurements of phyllaries were performed using light and scanning electron microscopy (SEM). Phyllaries are arranged in three or four circles. The analyzed species showed significant differences in the size of outer phyllary leaves. Among the examined species, the differences in the structure of phyllaries are mostly identified in the number of layers of hypodermis, organization of secretory tissues, position and number of vascular bundles. The SEM analysis showed that all examined species have non-glandular, uniseriate trichomes (NUT) surrounded by a rosette of 7 to 14 silicified cells. Many of examined species have NUT on both sides of leaves with the highest density on the abaxial surface and along the main veins. Also, few of them have NUT present only on the edge of phyllaries. Trichome cells have protuberances on cell walls, along their length. All examined species have presence of multicellular, linear, glandular trichomes (LGT), usually with the highest density on the abaxial surface of leaves. In some of the examined species, capitate glandular trichomes (CGT)

are present on the phyllaries, while species *H. mollis* and *H. nutalli* have CGT on both epidermal sides of the phyllaries. The obtained results significantly contribute to understanding morphological phyllary variability of wild *Helianthus* species and their potential use in breeding program of cultivated sunflower.

**ACKNOWLEDGMENTS:** This study was supported by Provincial Secretariat for Higher Education and Science of Vojvodina, project 142-451-2422/2018-03.

**KEYWORDS:** *Helianthus*, bracts, trichomes, morpho-anatomy.

Poster presentation 15 01 10

### ANATOMICAL INVESTIGATIONS OF EXTRAFLORAL NECTARIES OF *PRUNUS AVIUM* (L.) L. (ROSACEAE)

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Extrafloral nectaries are nectar-secreting plant glands developing on almost all aboveground plant organs except inside of flowers and are not involved in pollination. Contrary to floral nectaries that attract pollinators, extrafloral nectaries are attractive to predatory nectar-gathering insects, particularly some species of ants which feed on plant-eating insects, and thus significantly reduce the infestation levels and the leaf damage by herbivores. The aim of the present paper is to provide anatomical description of extrafloral nectaries occurring on leaf parts of *Prunus avium*. Fresh samples of full developed leaf glands were micromorphologically examined by stereomicroscope Nikon SMZ18. For light microscopy, the plant material was fixed in FAA and dehydrated with a graded ethanol series, infiltrated and embedded in paraffin, sectioned by sliding microtome (8–10  $\mu$ m thick) and stained with Safranin and Alcian blue. Observation and photographs were done using a light microscope Leica DM2000 equipped with a digital camera (Leica DFC320) and Leica IM1000 software. Extrafloral nectaries in *P. avium* occur at two positions: on both sides of the distal part of the petiole and throughout the leaf margin, being located in the top position on the teeth at the base of the leaf blade. The extrafloral nectaries on sides of the distal part of the petiole are larger than the marginal ones. Glands occurring in pairs are easily recognized, prominently elevated, intensively red-colored, and oval or discoid with a central concavity. Nectariferous tissue is composed of the two-layered secretory palisade-like epidermis and a multilayered subepidermal parenchyma vascularized by lateral veins derived from the leaf petiole. Vascular bundles composed of xylem and phloem are sometimes seen as nearly approaching the secretory epidermis. In parenchymatous tissue cells of irregular shapes and different sizes are distinguished: a subepidermal region with 3–4 layers of densely packed cells, and

an inner multilayered zone of cells with lignified walls and occasionally crystal druses inside.

**KEYWORDS:** *Prunus*, light microscopy, extrafloral nectaries

Poster presentation 16 01 11

### POLLEN MORPHOLOGY OF *MALABAILA AUREA* BOISS. (APIACEAE) - BALKAN ENDEMIC SPECIES

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*Malabaila aurea* Boiss. (Apiaceae) is Balkan endemic species distributed in Albania, Bulgaria, Greece, Montenegro, Macedonia, and Croatia, mostly growing on thermophilic and sunny habitats. Golden yellow flowers arranged in terminal compound umbels are frequently visited by insect pollinators. The pollen of *M. aurea*, collected from flowers at full flowering stage, has been examined by both, light microscopy and scanning electron microscopy (SEM) in order to contribute to taxonomic and melissopalynological research. For light microscopy the pollen grains were prepared according to the standard acetolysis method, mounted in glycerin jelly and observed with a Leica DM2000 microscope equipped with a digital camera (Leica DFC320) and Leica IM1000 software. For SEM study the pollen grains were observed using JEOL JSM-6390 LV electron microscope. Following palynomorphological features were examined: length of polar axis and equatorial diameter, size, shape, ornamentation, apertures, polarity, symmetry and exine thickness. The pollen grains of *M. aurea* are isopolar, radially symmetrical and at the interface between medium-sized and large. The ratio between the length of the polar axis ( $49.4 \pm 2.1 \mu\text{m}$ ) and the equatorial diameter ( $19.5 \pm 1.1 \mu\text{m}$ ) amounts  $2.5 \pm 0.2$  indicating prolate shape. The grains are equatorially constricted (bone-shaped) with obtuse polar caps, and in polar view they are triangular with interangular furrows. The grains are tricolporate with three straight sunken ectocolpi arranged regularly meridionally, of mean length  $28.5 \pm 2.2 \mu\text{m}$ , each with one endopore positioned in the indentations between the mesocolpial lobes. Mesocolpial width is  $9.9 \pm 1.6 \mu\text{m}$ . The sculpturing pattern is rugulate – microperforate. Exine is  $1.22 \pm 0.25 \mu\text{m}$  thick at the poles and twice as thick in equatorial region ( $2.59 \pm 0.49 \mu\text{m}$ ).

**KEYWORDS:** *Malabaila*, light microscopy, scanning electron microscopy, palynomorphology

Poster presentation 17 01 27

### QUALITY RELATED PERICARP ANATOMICAL CHARACTERISTICS OF PEPPER GENOTYPES

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Anatomical characteristics affect technological quality and use value of edible fruits. Pepper is one of the most important vegetable species due to its high nutritional value, as well as due to its multiple uses in human nutrition (like fresh consumption, cooking, pickling) and in industrial processing. Quality requirements and desirable traits depend on the final use of fruits. Anatomical characteristics of fruits of 12 pepper genotypes selected for different purposes (Amfora, Piquillo de Lodosa, Kurtovska kapija, Kalifornijsko čudo - California Wonder, Anita, Šorokšari, Novosađanka, 214/14, Zeleni rotund, HS-2, HS-6, Venčarka) were analyzed. Cross sections of ripe fruits were hand-made and analyzed using light microscopy. Observations and measurements of pericarp parameters were performed using Image Analyzing System MotiC. Pepper fruit pericarp was composed of one-layered epidermis, 1-5 layers of collenchyma, vascular bundles and well developed parenchyma, with huge vesicular cells adjacent to endocarp. Differences among the genotypes were recorded in quantitative anatomical parameters. The thickest pericarp was recorded in peppers grown for fresh consumption (Kapia and Bell peppers) and pickling (Tomato peppers). Mesocarp was the thickest and with most numerous cell layers in tomato peppers, which had less developed peripheral tissues and the smallest proportion of cuticle, exocarp, endocarp and collenchyma tissue. The thinnest pericarp had peppers grown for spices, which were also characterized by the higher percentage of peripheral tissues (cuticle, exocarp, and collenchyma), thicker endocarp and lower mesocarp thickness. Discriminant analysis of the anatomical characteristics of the fruits showed a clear separation of the groups of genotypes according to their usage, although none of the analyzed parameters contributed significantly to this discrimination. The findings confirmed that anatomical parameters had an important role in use value determination of pepper fruits.

**KEYWORDS:** pepper fruit, pericarp anatomy, mesocarp

Poster presentation 18 01 26

### NUTLET ANATOMY, MICROMORPHOLOGY AND RAMAN SPECTROSCOPY OF THREE MACEDONIAN *SALVIA* SPECIES

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*Salvia* L. (sage) is the largest genus of Lamiaceae family comprising nearly 1000 species of which 36 species is represented in Europe and 15 species in Macedonia. *Salvia amplexicaulis* Lam. is distributed in the Balkan Peninsula, *S. ringens* Sibth. & Sm. is distributed in Eastern part of the Balkan Peninsula, while *S. jurisicii* is endemic species of the central part of the Republic of Macedonia. Mericarps (nutlets) of *Salvia amplexicaulis*, *S. jurisicii* and *S. ringens* were collected during fruiting phase in several localities in Macedonia and subjected to microscopical analyses for the first time. The objective of this study was structural, micromorphological and spectroscopic investigation of nutlets and mucilage of the mentioned *Salvia* species. Light microscopy (LM) and scanning electron microscopy (SEM) were employed in order to examine nutlets and mucilage morphology, as well as nutlets micromorphology and anatomy, while Raman and Attenuated Total Reflectance Fourier Transform Infrared (ATR FT-IR) spectroscopy were applied to obtain data on their chemical composition. The nutlets of *S. amplexicaulis* are the smallest ones and of prolate-spheroidal shape, while in *S. jurisicii* and *S. ringens* the shape is spherical. The largest nutlets are those of *S. ringens* (3.08 mm in length and 2.27 mm in width). Nutlets were pale to dark brown colored. Abscission scar was triangular in *S. amplexicaulis* and round in other two species. Trichomes were absent and surface ornamentation was reticulate in all examined species. Myxocarpy was obtained after 15 minutes of wetting in *S. amplexicaulis* and *S. jurisicii*, while larger nutlets of *S. ringens* produced mucilage after 45 minutes. Pericarp cross sections of all examined *Salvia* species showed considerable uniformity from anatomical point of view. Mucilage in all studied species was yellowish and transparent containing fibrils, except in *S. ringens*. The analysis of spectra obtained by Raman spectroscopy revealed that the nutlets are predominantly composed of  $\alpha$ -linolenic and linoleic acids. The results from ATR FT-IR spectroscopy showed that mucilages primarily consisted of polysaccharides. In conclusion, these findings provide contribution to the knowledge on the genus *Salvia* which could be useful in taxonomical investigation of this genus.

**KEYWORDS:** *Salvia*, nutlets, mucilage, micromorphology, microscopy, spectroscopy

Poster presentation 19 01 14

### CARPOLOGICAL INVESTIGATIONS ON SOME *CARDUEAE* SPECIES (ASTERACEAE) FROM TURKEY

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The tribe *Cardueae* comprises more or less 500 taxa included 39 genera in Turkey, according to recent reports. In the present study, cypsela morphological and anatomical structures of six representative species, two of them are endemic to Turkey, belonging to six genera were characterized in detail by using stereomicroscope and light microscope. The species were evaluated comparatively in the aspect of carpological variations and their anatomies were presented in here for the first time, except for *Xeranthemum annuum*. Morphological features including size, shape and color of cypselas were examined. Cypselas colors differ from dark brown-blackish to stramineous. Their shapes changes are oblong, oblong-cylindrical, widely oblong and narrowly oblong. The smallest cypselas occurred in *X. annuum*, the biggest one was found in *Echinops orientalis*, and the widest fruit was detected in *Arctium platylepis* among the investigated taxa. From anatomical observations, pericarp anatomical structures were described, as well as the structure of testa. It was determined that *Arctium platylepis* has parenchymatic pericarp structure, while *Psephellus hypoleucus* has sclerenchymatic one. On the other hand, both parenchymatic and sclerenchymatic cells were observed in *Xeranthemum annuum*. Two layers of endosperm were determined in *Echinops orientalis*, differently from other species. Tetragonal crystals were observed in three species (*Arctium platylepis*, *Onopordum turcicum* and *Psephellus hypoleucus*). Data obtained from this study were compared with the data present in literature. These results demonstrate that the compared fruit morphological and anatomical characters among species are significantly different and can be used as taxonomic markers in their classifications.

**KEYWORDS:** *Cardueae*, cypselas, anatomy, Turkey

Poster presentation 20 01 04

### COMPARATIVE PERICARP ANATOMY OF WILD PERENNIAL *HELIANTHUS* L. SPECIES

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The production of high yielding cultivated sunflower genotypes, which are of higher and improved quality, as well as of increased resistance to abiotic and biotic factors, has been the main goal in the process of breeding. Apart from the taxonomically useful information, the characteristics of pericarp are very important in defining the fruit yield potential. General anatomy of the sunflower fruit is relatively well known, but not enough attention has been given to the research of the anatomic characteristics of the pericarp as potentially important parameters in the breeding programme. Therefore, we considered that it was important to make very detailed comparative analysis of pericarp anatomic characteristics of the 19 wild perennial *Helianthus* species. The plant material was cultivated in the experimental field. For anatomical observation mature achenes were randomly selected and cross sections were obtained from the middle part of the fruit, using cryotechnic procedure. Observations and measurements of the pericarp features were carried out using the scanning electron microscope (SEM) and light microscope with Image Analyzing System. The results obtained indicated the presence of some quantitative and qualitative differences in the pericarp anatomical characteristics among analyzed species. SEM analysis of the pericarp epidermis shows that cuticle varies in texture. Among the examined species the differences in the structure of pericarp are mostly recognized in the number of layers of hypodermis and sclerenchyma, by the percentage of sclerenchyma and in the shape of sclerenchyma groups. The results of the Multivariate Discriminant Analysis showed that species *Helianthus giganteus* L., *H. resinosus* Small, *H. hirsutus* Raf. and *H. glaucophyllus* D.M.Sm had remarkably bigger tissue differentiation than other species. Variations of the pericarp anatomy among the wild perennial *Helianthus* species could be related to their different dispersal strategies, as well as different dormancy and germination requirements.

**ACKNOWLEDGMENTS:** This study was supported by Provincial Secretariat for Higher Education and Science of Vojvodina, project 142-451-2422/2018-03.

**KEYWORDS:** anatomy, pericarp, sunflower, wild perennial

Poster presentation 21 01 24

### EFFICIENT METHOD FOR *HYSSOPUS OFFICINALIS* L. MICROPROPAGATION

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In the case of medicinal plants, micropropagation procedures are nowadays used for obtaining a large number of valuable individuals starting from a donor plant with high production capacity. A highly efficient and low-cost protocol for tissue culture introduction and micropropagation of the medicinal plant hyssop (*Hyssopus officinalis* L.) has been conducted. Stem tips from plants vigorously growing under controlled conditions were used as initial explants. The explants were sterilized with 0.04% mercuric chloride (HgCl<sub>2</sub>) solution for 20 minutes and washing three times with sterile distilled water in 15 minutes. The cultural media was full and half strength Murashige and Skoog (MS) medium containing indole-3-butyric acid. Full and ½ MS media without auxin were used as controls. For each variant, 20 glass tubes with two plants were used. In each tube, two tip and nodal explants were grown. Maximum shoot and root number were obtained on ½ MS medium supplemented with 0.1 mg L<sup>-1</sup> indole-3-butyric acid at the same time after four weeks of culture. The number of shoots per explant and shoot height were considered. The data on rooting percentage, the number of roots per plant and root length were collected after the same cultural period. The highest percentage of survival 85% for this medicinal plant was recorded in mixture of soil, sand and perlite (2:1:1 v/v/v). This mixture was most suitable for acclimatization of all propagated plants. Ex vitro acclimatization was carried out at 24°C and 70% relative humidity under 16 h illuminations (50 µmol m<sup>-2</sup>s<sup>-1</sup>). After an adaptation period, the plants were transferred to the field. Developed in vitro techniques could provide a promising way for successful large-scale propagation of *Hyssopus officinalis* L.

**ACKNOWLEDGEMENTS:** This study was conducted with financial support from National Science Fund at the Bulgarian Ministry of Education and Science, Bilateral Joint Research Project DNTS Slovakia 01/3, 2016.

**KEYWORDS:** *Hyssopus officinalis* L., micropropagation, acclimatization, indole-3-butyric acid

Poster presentation 22 01 36

### EFFECT OF GRAFTING ON WATERMELON ANATOMY AND GENETICS

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Grafting is an important, ancient plant propagation practice, widely used with fruit trees and in recent decades also with herbaceous plants. Grafting can contribute to an increased resistance to abiotic and biotic stresses as well as to higher yields. The aim of the present study was to investigate the changes in anatomical and genetic traits of the scion, induced by grafting watermelon (*Citrullus lanatus*) on calabash (*Lagenaria siceraria*) rootstock. Plant lines 'Lady' (*C. lanatus*) and 'Argentario' (*L. siceraria*) were grown in Hungary, Cece. 15-15 stem and leaf samples per population were collected from healthy, well-developed plants for microtome sectioning. The qualitative and quantitative characters of the transections of grafted and ungrafted *C. lanatus* were compared. For exploratory genetic analysis, Random Amplified Polymorphic DNA (RAPD) was used. The leaf extracts of the samples were tested by 23 primers, from which 8 were suitable for further analysis. Statistical analysis of anatomy was performed using F-test. Rho values and Principal Coordinates Analysis (PCoA) of the RAPD fragment patterns were calculated by R environment 3.1.2. software. Significant differences were found in the thickness of the one layered epidermis and in the characters of the vascular bundles in the stem. There were 8 vascular bundles arranged in one ring in ungrafted 'Lady', while 10 bundles in two rings in grafted ones. The xylem was significantly thicker in the grafted lines than in the ungrafted samples, because of the higher diameter of the xylem vessels. The only significant difference in leaf anatomy was the size of the rib above the main vascular bundle, the grafted samples having thicker midrib region than the ungrafted plants. Segregation of the samples estimated by Rho values showed that the rootstock was well separated from grafted (0.6051) and ungrafted (0.6367) 'Lady'. Furthermore, the grafted and ungrafted plants can be distinguished based on their RAPD fingerprint (0.2055). Based on our research, grafting influenced significantly the genetic background, and the process may provide i.a. better water uptake and transport for the scion, as supported by anatomical observations.

**KEYWORDS:** *Citrullus lanatus*, leaf, stem, RAPD, anatomy, grafting

Poster presentation 23 01 34

### CHARACTERISTICS OF THE REPRODUCTIVE FEATURES OF *SATUREJA PILOSA* VELEN. VAR. *PILOSA* (LAMIACEAE)

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The genus *Satureja* L. (Lamiaceae) comprises about 200 species, largely distributed in the Mediterranean region of Europe, and in West Asia, North Africa and South America. Five of the *Satureja* species are found in the Bulgarian flora. The objective of the present work was to conduct the first embryological study of the Balkan endemic *S. pilosa* Velen. var. *pilosa* from the Bulgarian flora. The study established the features of the male and female reproductive sphere of the species in order to reveal the mode of reproduction, character, size and state of its populations. The plant material collected from plants of natural populations above the town of Sopot in Stara Planina Mountains was fixed in a mix of FAA and treated according to the Classical Paraffin Method. As a result, the structure in the male and female generative sphere and the processes of gamete development, pollination, endosperm and embryo formation were revealed. The male generative sphere was: tetrasporangiate anthers, Dicotyledonous-type of development of the anther wall that consists of epidermis, fibrous endothecium, one middle layer and glandular tapetum forming placetoids at the stage of one-nucleate pollen, predominantly tetrahedral microspore tetrads formation in result of simultaneous microsporogenesis, 2-celled mature pollen grains. The female generative sphere was: anatropous ovule, unicellular archesporium, Polygonum-type of development of the female gametophyte, Onagrad-type of embryogenesis. The features of the embryological structures and processes, and absence of apomixis characterize *S. pilosa* as a sexually reproducing species. The observed normal pass of processes in the generative sphere provides a high reproductive potential of the species and stability in the population numbers. The balanced processes and stable structures, and only sexual reproduction may decrease the adaptability of the species to environmental conditions. The endemism of *S. pilosa* is probably due to its dependence on specific environmental conditions, and more likely is a consequence of the low plasticity of the generative sphere. Therefore, measures such as conservation of its habitats and limitation of anthropogenic pressure on the populations would be most efficient for the preservation of this species.

**ACKNOWLEDGEMENTS:** This study was supported by the Agricultural University Plovdiv, Project № 11-18.

**KEYWORDS:** *Satureja*, embryology, male and female gametophyte, embryo, endosperm

Poster presentation 24 01 41

## GERMINATION AND GROWTH OF WHEAT CULTIVARS UNDER PEG-INDUCED DROUGHT

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Soil water potential is an important environmental factor influencing germination and seedling development, and drought stress is one of the most significant abiotic constraints of growth and productivity of crop plants worldwide. The main purpose of this study was to define the influence of reduced water potential on germination indicators and physiological and biochemical parameters of seedlings of two winter wheat cultivars (Felix and El Nino, with moderate and low drought resistance). To imitate the water potential of the actual conditions in the soil, eight different concentrations of polyethylene glycol (PEG 6000) were used, ranging from -0.03 (field capacity) to -1.5 MPa (permanent wilting point). The variables measured were final germination percentage, mean germination time and germination rate index, fresh weight of seedlings, and shoot and root length, as well as lipid peroxidation (as malondialdehyde content), osmotic substances (soluble proteins and free proline) and the activities of scavenging enzymes (guaiacol peroxidase, ascorbate peroxidase and catalase) in both shoots and roots of seedlings. The results indicated that reduced water potential decreased germination indicators and seedling vigor in both cultivars. The cultivar El Nino had a slightly lower germination percentage in the control growth medium (89%), unlike to Felix with 97%, as well as higher mean germination time compared to Felix. Germination index of El Nino significantly decreased only at the water potential of germination substrate at -0.7 MPa, while germination index of Felix significantly decreased already at -0.3 MPa. Root length decreased in both cultivars by 30%, and shoot length by 16% in El Nino and 46% in Felix cultivars. With increasing PEG concentrations, two times more proline accumulated in El Nino seedlings compared to Felix, and in both cultivars roots accumulated twice as more proline than shoots. Lipid peroxidation and concentrations of antioxidative enzymes increased, more in Felix than El Nino, indicating that plants sustained considerable damage and that damage was proportional to water shortage, but the accumulation of osmolytes and increased activity of antioxidative enzymes helps them cope with drought injury.

**KEYWORDS:** drought, PEG 6000, wheat seedlings, germination, proline content, antioxidative enzymes

Poster presentation 25 01 47

## GERMINATION AND VIABILITY OF SEEDS OF TANACETUM CINERARIIFOLIUM (ASTERACEAE)

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*Tanacetum cinerariifolium* (Trevir.) Sch. Bip. is a perennial herbaceous plant. The species is endemic to the east Adriatic coast. The seeds of *T. cinerariifolium* are oblong, ribbed, cylindrical with 5 to 7 ridges, cuneiform, 3-5 mm long and 0.8-1.0 mm thick, and with a slightly lustrous surface. It is believed that the use of pyrethrins in insecticide preparations dates back to Persia, where *Pyrethrum roseum* Bieb. and *Pyrethrum garneum* Bieb. have been used since ancient times. The insecticidal properties of *T. cinerariifolium* were discovered later. Natural pyrethrins have some of the qualities of the ideal pesticide and constitute one of the most important insecticides. They are biodegradable, very effective against a wide range of insects acting as a contact insecticide, and the probability that resistance will develop is low. The purpose of this study is to determine the effect of different seed storage periods on seed germination and viability. Seeds from *ex situ* collection of pyrethrum in Bulgaria were used as a starting material (freshly collected mature seeds, seeds stored for 1 year, for 2 and for 3 years). Seed germination was tested on filter paper in petri dishes and *in vitro* on basal MS medium, for a period of 8 weeks, 4 repetitions 100 seeds each one. An equal number of freshly collected mature seeds, and seeds stored for 1, 2 and 3 years (4x100) were used for viability determination following tetrazolium test. About 2/3 of the seeds were empty. The percentage of viable seeds was highest for the freshly collected mature seeds (22.25%) and viability declined almost twice after one year storage (13.25%), while no viable seeds were found after 3 years. Germination of freshly collected seeds began in the first week, but with the increase of the storage duration some delay of the germination start was observed. Germination rate was 20.75% for seeds in petri dishes and 17.75% on MS medium ( $p < 0.05$ ), and both germination time and germination period were longer *in vitro*.

**ACKNOWLEDGEMENTS:** The authors are grateful to the Program for Support of Young Researchers and PhD Students at the Bulgarian Academy of Sciences (Grant no. DFNP - 17-109/28.07.2017) for the financial support.

**KEYWORDS:** Dalmatian pyrethrum, *Pyrethrum cinerariafolium* Trev., *Chrysanthemum cinerariafolium* Bocc, seed germination, seed viability

Poster presentation 26 01 43

## ANATOMICAL CHARACTERISTICS OF LEAVES AND PRESENCE OF MYCORRHYZA IN THREE BORNMUELLERA SPECIES (CRUCIFERAE) FROM THE BALKAN PENINSULA

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The genus *Bornmuellera* Hausskn. is represented by three species in Europe: *B. baldaccii* (Degen) Heywood from Albania and Greece, *B. dieckii* Degen from Mt. Ostrovica in Kosovo, and *B. tymphaea* (Hausskn.) Hausskn. from N Greece. All three species thrive on serpentine habitats characterized by unfavorable Ca/Mg ratio, low level of nutrients, and high level of heavy metals, warm summer and low water content in the soil. However, they successfully survive in these hostile habitats. One reason for this is that they are hyperaccumulators of nickel with levels of this metal in their tissues being above 1000 µg/g of dry weight and even reaching maximum values of 30000 µg/g dw. But, what other characteristics helps them survive in this environment? To begin answering this question, we examined anatomical characteristics of their leaves and investigated presence of mycorrhiza on their roots. Three European *Bornmuellera* species have distinguished xeromorphic leaf characteristics. In *B. tymphaea* leaves are very densely covered by bifid hairs on both sides. Leaves are sparsely pubescent in *B. baldaccii* with appressed, bifid to 6-fid hairs more present on the lower than on the upper leaf side, while bifid trichomes are scattered on the lower and sporadic on the upper leaf side of *B. dieckii*. Sporadic glandular hairs can be found mostly on the margin of the leaves or on the lower epidermis. All three species have thick cuticle on the leaf surface, and amphistomatous leaves. Leaves are isolateral. Mesophyll consists of densely packed cells, and it is characterized by a domination of palisade tissue. Vascular bundles are numerous and surrounded by parenchyma cells. All these anatomical features of the leaves points to their maximal adaptation to drought and high insolation in the habitat. Vesicular arbuscular mycorrhizal fungi (VAM) are found in roots of all three investigated *Bornmuellera* species suggesting its possible role in a better supply of water and nutrients from dry and poor soil to the plant.

**KEYWORDS:** serpentine, nickel, hyperaccumulators, anatomy, trichomes, hairs

Poster presentation 27 01 48

## PECULARITIES OF THE REPRODUCTIVE BIOLOGY OF ALKANNA TINCTORIA (BORAGINACEAE)

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*Alkanna tinctoria* (L.) Tausch is a historical plant that has been used for the treatment of macular eruptions and infectious diseases, such as measles, sore-throat, carbuncles and burns, and has also been used as a dye in the cosmetic and fabric industries. In Bulgaria the species has a limited distribution and small populations due to the erosion, collecting for medicinal use and the grazing and trampling by livestock. Therefore, it is included in the Red Book and Red List of Higher Plants and is protected by the Biodiversity Act. The present investigation was carried out to reveal the peculiarities of the male and female gametophyte of *A. tinctoria* in connection with the estimation of its reproductive potential that directly affects the size of its populations. For this purpose, flowers and flower buds in different developmental stages collected from plants of two natural population of *A. tinctoria* in the Bulgarian flora (village Lesovo and village Harsevo in Southwest Bulgaria) was fixed in the mix of FAA and threated according the classical paraffin methods. As a result the main features of the structures and processes in the male and female generative sphere were established. They determined *A. tinctoria* as a sexually reproducing species. The produced in result of development of the male gametophyte mature pollen grains was morphologically uniform and fertile in high degree. The processes leading to formation of the embryo passed normally that is a prerequisite to formation of viable seeds. The high amount of fertile pollen, effective seeds formed, and the stability of structures and processes in the reproductive sphere guarantee the stability of size of the populations of *A. tinctoria*.

**KEYWORDS:** generative sphere, embryology, male and female gametophyte, embryo

Poster presentation 28 01 50

### WITHIN-PLANT VARIATION OF DEVELOPMENTAL INSTABILITY IN MALE AND FEMALE *MERCURIALIS PERENNIS* IN CONTRASTING LIGHT HABITATS

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Plants, as sessile organisms, are exposed to macro- and microenvironmental changes, the suboptimal stress conditions leading to developmental instability that can be quantified by assessing the fluctuating asymmetry (FA) of organs or their parts. The analysis of repeated plant organs (such as leaves of successive nodes) can be used to assess the intraindividual aspect of developmental instability patterns. In dioecious species, gender of plant can also be one of the factors affecting asymmetry levels. The aim of this study was to explore the inter- and intra-individual patterns of developmental instability in natural populations of a dioecious species in contrasting light habitats, with special focus on within-plant asymmetry patterns. Male and female individuals of dioecious understory forb *Mercurialis perennis* (Euphorbiaceae) were sampled from two contrasting light habitats, shaded (spruce forest) and open (sun-exposed field), located at mountain Kopaonik. Four leaf traits were measured: petiole length, midvein length, leaf blade width and leaf blade surface area. Leaf measurements were used to calculate univariate, as well as multivariate, size-scaled indices of fluctuating asymmetry for each node. Nested analysis of variance, with habitat, sex and node as fixed factors, and individual as random factor, was used to investigate the inter- and intra-individual variation in FA indices. Our results showed that the highest values of FA (i.e. developmental instability) have been found for the petiole length, while leaf blade width and midvein length had lower values of FA. We discussed the higher variability and developmental instability of petiole length in context of the role of this trait in leaf orientation. We found no significant effects of plant gender, light habitat and habitat x gender interaction on the analyzed FA indicators. On the other hand, between and within-plant variations in FA levels were significant, indicating the significant effects of small scale spatial and temporal variation of light conditions for plant development and emphasizing the importance of microenvironmental conditions.

**KEYWORDS:** developmental instability, fluctuating asymmetry, plant sexual dimorphism

Poster presentation 29 01 17

### EFFECTS OF LOW AND HIGH TEMPERATURE STRESSES ON POLLEN GERMINATION AND POLLEN TUBE GROWTH IN *CORYLUS AVELLANA* CULTIVARS

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Pollination is one of the most important factors in management of hazel orchards in order to obtain a good yield. At the time of pollination, ovary is not formed and grows only if the flower is pollinated. After compatible pollination, pollen grains wait for formation of the ovary. Pollen germination and tube growth may be exposed to different temperature stresses due to extreme changes of day and night temperature or seasonal temperature changes during the long waiting process. And this is one of the important factors affecting yield. For this purpose, we analyzed the effects of low and high temperature stresses on the pollen germination and pollen tube growth by focusing on the callose distribution and actin organization in four Turkish *Corylus avellana* cultivars, Sarı, Palaz, Kara, Yomra. Pollen grains were stressed by low (4 °C) and high temperature (30 °C) treatment for 24 h using incubation chambers. Control pollen grains were kept at room temperature during the treatment. Both, control and stressed pollen grains were germinated in BK medium supplemented with 12 % sucrose. Germination rates and pollen tube lengths of all cultivars are highest at 20 °C and decrease with low and high temperature stresses. In vitro pollen germination and tube length of Kara cv. are more susceptible to low and high temperature stresses, while Yomra cv. is more resistant. Dense callose accumulation is observed on the tube tips of Kara cv. at stressed groups while accumulation is not observed on the tube tips of other cultivars. Pollen tubes show some abnormalities such as swelling and thinning at stressed groups for all cultivars, but more at 30 °C. Also, actin organizations of pollen tubes show major disruption at stressed groups for all cultivars. However, the most devastating effect is found at 30 °C. Actin became disorganized in the shanks of the pollen tubes, particularly in the balloon-shaped tip, causing broadening of the pollen tube shank. Results can provide useful information for optimizing pollination, choosing pollinator and increasing the fruit set, by detecting the temperature sensitive and tolerant cultivars.

**KEYWORDS:** actin cytoskeleton, callose, *Corylus avellana*, pollen germination, pollen tube, temperature stresses

Poster presentation 30 01 25

### EFFECT OF ARBUSCULAR MYCORRHIZAL FUNGI ON THE SOIL ENZYMES AND ESSENTIAL OILS CONTENT OF IN VITRO PROPAGATED *HYSSOPUS OFFICINALIS* PLANTS

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*Hyssopus officinalis* L., Lamiaceae, commonly called hyssop, is an aromatic, semi-evergreen, woody-based, shrubby perennial plant. Hyssop is a good expectorant and antiviral herb commonly used to treat respiratory conditions such as influenza, sinus infections, colds, and bronchitis. Most of its medicinal properties are attributed to the essential oil of hyssop. The arbuscular mycorrhizal fungi (AMF) inoculation is a promising technology in the sustainable agricultural system to improve the plant productivity performance. AMF can increase plant growth, photosynthesis, nutrients storage, metabolites and beneficial chemical compounds and decrease soil borne plant diseases by inhibition of fungal pathogen. Several studies have shown different effects on secondary metabolites, biomass production, as well as root and soil enzyme activities. The successfully adapted to field conditions in vitro plants were inoculated with arbuscular mycorrhizal fungi (*Claroideoglossum claroideum*, ref. EEZ 54). Soil enzyme activities are often used as indices of microbial activity in the soil. The plants flowered within three months after transplantation. All the in vitro propagated hyssop plants displayed normal development under the field conditions. It was established that the activities of acid and alkaline phosphatases in root and soil were significantly higher when plants are inoculated with *Claroideoglossum claroideum* in compare with non-inoculated plants. Enhanced levels of soil enzymes are in correspondence with the level of colonization the hyssop roots with AM fungi 84%. Mycorrhizal inoculation enhanced the levels of easy extractable and total extractable glomalin – related proteins. Glomalin, a glycoprotein of AMF origin, has been proposed as a soil particle binding agent and assist in increasing water and nutrient uptakes According to plant essential oil content, there was no significant difference between non-inoculated and inoculated hyssop plants. The beneficial effect of mycorrhizal fungi on the soil enzymes, biologically active compounds in aerial plant parts are important to support the development of organic and nonpolluting agriculture.

**ACKNOWLEDGMENTS:** This study was conducted with financial support from National Science Fund at the Bulgarian Ministry of Education and Science, Bilateral Joint Research Project DNTS Slovakia 01/3, 2016.

**KEYWORDS:** *Hyssopus officinalis*, micropropagation, acid and alkaline phosphatases, glomalin, essential oil

Poster presentation 31 01 15

### EFFECTS OF A TOXIC MICROCYSTIS (CYANOBACTERIA) STRAIN ON ROOT CELLS OF RICE

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Cyanobacteria constitute an ancient group of organisms of great ecological significance, present in a wide variety of aquatic habitats. Cyanobacteria produce a wide range of bioactive compounds, many of which, such as microcystins, are considered as hazardous for other organisms, including plants. Rice (*Oryza sativa*) is a crop species of great commercial value worldwide, its cultivation being closely related to the aquatic environment and, subsequently, prone to all the hazards due to the occurrence of cyanobacteria in the water surrounding the plants. The present study aims to investigate the impact of cyanotoxin-producing cyanobacteria on plant cells. The effects of the extract from a toxic (microcystin-producing) freshwater cyanobacterial strain (*Microcystis flos-aquae* TAU-MAC 1510), isolated from Lake Pamvotis, Greece, on four-day old rice seedlings were studied. The seedlings were exposed to the extract for various time periods. Tubulin immunolabeling and DNA staining were then performed on root tip cells, which were observed by fluorescence microscopy, while other root tips were fixed and prepared for transmission electron microscopy. Some root tips were stained for detection of either hydrogen peroxide (H<sub>2</sub>O<sub>2</sub>) or presence of dead cells and observed by light microscopy. Microtubules were disassembled in a time-dependent way, an effect that has been associated with the inhibition of protein phosphatases 1 and 2A by microcystins. Chromatin was also abnormally condensed, and this has been related to disturbances of histone phosphorylation in mitotic cells. Moreover, ultrastructural alterations in membranes and organelles were linked to the detected oxidative stress caused by the toxic extract. This oxidative stress seems to result in cell death, as confirmed by Evans Blue staining, probably via the programmed cell death pathway, in a progressive and time-dependent process.

**KEYWORDS:** cyanobacteria, microcystin, microtubule, oxidative stress, plant cell, programmed cell death

### VARIATIONS OF CHLOROPHYLL A FLUORESCENCE INDUCED BY CLIMATE CHANGES IN WINTER BARLEY GENOTYPES

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Plant adaptation to climate change presents one of the major issue today. The insight into complex networks of molecular interactions controlling plant acclimation to field conditions could significantly improve chances of developing crops with enhanced tolerance to field conditions. To develop plants tolerant to a wide range of stresses, new approaches are needed to investigate the response of crops to a combination of different abiotic stresses. The main goal of breeding process is to select barley cultivars with better adaptation to diverse climate regimes. In the present study we evaluated photosynthetic performance in ten selected winter barley genotypes responding to variations in environmental conditions during growth season. To evaluate condition of the photosynthetic apparatus, fast chlorophyll a fluorescence was measured in the field at three developmental stages: booting, anthesis and early grain filling. Furthermore, normalizations and subtractions of polyphasic fluorescence transients (OJIP) were made to reveal K and L-band. Lower amount of precipitation during anthesis induced instability of photosystem II (PSII) observed as positive K-band in six and positive L-band in seven genotypes. Positive inflections of these bands suggested slightly destabilization of oxygen evolving center and light induced antenna dissociation and lower PSII stability, respectively. At grain filling all genotypes displayed negative K and L-bands, suggesting increase of stability within PSII, as well as high energetic connectivity of PSII. However, the performance index (PIabs) continuously increased from booting to grain filling in most genotypes. At anthesis this was mainly influenced by the increase of electron transport beyond primary electron acceptor QA, while at grain filling it was attributed to the increase of density of reaction centers per chlorophyll basis. As the response to mild drought stress during anthesis, two types of barley genotypes can be distinguished: tolerant and susceptible.

**KEYWORDS:** *Hordeum vulgare* L., fluorescence, performance index, L-band, K-band, OJIP

### PHOTOSYNTHETIC CHARACTERISTICS OF DOG ROSE (*ROSA CANINA* L.), YARROW (*ACHILLEA MILLEFOLIUM* L.), DANDELION (*TARAXACUM OFFICINALE* WEBER.) AND BLACKBERRY (*RUBUS CAESIUS* L.) IN RELATION TO SOIL CONTAMINATION WITH CADMIUM AND LEAD

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Soil contamination by heavy metals is a global-scale problem, due to their mobility in the soil-plant system and toxicity to most living organisms. Cadmium (Cd) and lead (Pb) are potent and widespread pollutants, and they are not essential for plant metabolism. However, plants are able to absorb these elements from the growth substrate, and to accumulate them within vegetative and/or reproductive organs at various levels. The aim of our investigation was to define photosynthetic characteristics and gas-exchange parameters (net photosynthesis, transpiration, stomatal conductance, substomatal concentration of CO<sub>2</sub>, chlorophylls and carotenoids concentration) of blackberry, dandelion, dog rose and yarrow, in relation to Cd and Pb-soil contamination. Gas-exchange parameters were measured in vivo, and concentration of photosynthetic pigments was determined in acetone extracts of leaves collected from both contaminated and uncontaminated sites in the Vojvodina province (Republic of Serbia). Rate of photosynthesis was lowered in dandelion at contaminated site, with respect to uncontaminated one. Heavy metal contamination did not considerably affect photosynthetic rate and transpiration in yarrow and dog rose plants. Despite the negative influence of soil contamination upon chlorophylls and carotenoids level in blackberry, rate of photosynthesis, transpiration and stomatal conductivity were increased at contaminated site. These results could be attributed to some favorable conditions at the site, such as light intensity. In most cases, concentration of chlorophylls and carotenoids was considerably affected by soil contamination. The pigments were considerably decreased in blackberry and dandelion plants at contaminated sites, while their concentration tended to increase in dog rose and yarrow. Obtained results indicated various physiological responses of studied species in relation to increased Cd and Pb in soil. Inconsistent changes of physiological parameters analyzed in this work could be explained by differences in heavy metal uptake, accumulation, as well as different physiological defense mechanisms involved in maintenance of stable metabolism under excessive Cd and Pb in soil.

**ACKNOWLEDGEMENTS:** Research was conducted within the project titled: "Biologically active components and medical potential of functional food grown in Vojvodina Province, Serbia"

no. 114-451-2149/2016-0, financed by government of Province of Vojvodina.

**KEYWORDS:** photosynthesis, chlorophylls, carotenoids, heavy metals, soil pollution

### THE EFFECTS OF BISPHENOL A ON THE PHOTOSYNTHETIC MACHINERY OF THE SEAGRASS *CYMODOCEA NODOSA*

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The xenoestrogenbisphenol A (BPA) is an intermediate chemical used for synthesizing polycarbonate plastics and is an emerging organic pollutant of environmental concern. Classified as "moderately toxic" and "toxic", it causes adverse effects on aquatic biota. Although information about BPA toxicity on aquatic fauna is available, data on aquatic flora remain scarce, in particular for marine angiosperms, among which is *Cymodocea nodosa*. Considering the above, in the present study we investigated the effects of environmentally relevant BPA concentrations (0.3, 1 and 3 µg L<sup>-1</sup>) on the photosynthetic machinery of *C. nodosa* for 1, 2, 4, 6, and 8 days. BPA toxicity effects were higher at 3 µg L<sup>-1</sup> BPA exerting their deleterious effects faster (i.e. from the first day). Generally, a "mosaic" type BPA effect pattern was observed, with "unaffected" leaf areas and areas with intense negative effects. In particular, as revealed by DIC and CLSM microscopy, the negatively affected leaf areas lost their chlorophyll auto-fluorescence, while the cells appeared filled with dense material, accumulating H<sub>2</sub>O<sub>2</sub> in a time-dependent manner, as revealed by DCF-DA staining. Ultrastructural defects were also observed. At even relatively short BPA-treatments (4 days) with 3 µg L<sup>-1</sup>, plastids had damaged thylakoids appearing electronically dense, while effects on Golgi dictyosomes and the endoplasmic reticulum were also observed. In the "unaffected" leaf areas cells exhibited increased phenolic compound production, as revealed by K<sub>2</sub>Cr<sub>2</sub>O<sub>7</sub> staining. Chlorophyll fluorescence imaging analysis confirmed the "mosaic" type effect pattern with reduced number of open photosystem II (PSII) reaction centers (qp) and lower electron transport ratio (ETR) in the "affected" leaf areas. However, there was no effect on the quantum yield of non-regulated non-photochemical energy loss in PSII (ΦNO), which consists of chlorophyll fluorescence internal conversions that lead to the formation of singlet oxygen via the triplet state of chlorophyll, with the exception in 3 µg L<sup>-1</sup> BPA treatments that resulted to a significant high singlet oxygen production. Bisphenol A, at environmentally relevant concentrations, caused defects with

the upregulated H<sub>2</sub>O<sub>2</sub> production to be a key factor causing both oxidative damage but also triggering retrograde signaling conferring tolerance to BPA in the "unaffected" leaf areas.

**ACKNOWLEDGEMENTS:** Funds of MIS 5001552, operational programme "Human resources development, Training and Lifelong Learning, ESPA (2014-2020)"

**KEYWORDS:** chlorophyll fluorescence, golgi dictyosomes, hydrogen peroxide, marine angiosperm, thylakoids, xenoestrogen

### EVALUATING PHOTOSYNTHETIC PERFORMANCE OF DIFFERENT BARLEY CULTIVARS UNDER STRESS USING CHLOROPHYLL A FLUORESCENCE

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The current cereal production is challenged by the abiotic stress induced by climate changes. Selection and breeding processes aim to select barley cultivars with better adaptation to stress. Drought stress presents one of the environmental stresses with significant impact on agricultural production worldwide. The main objective of this study was to screen and evaluate twenty winter barley cultivars for their photosynthetic performance by using fast chlorophyll a fluorescence parameter during growth season in the experimental fields where plants were exposed to stressful conditions due to drought treatments. Measurements were taken at three growth stages: booting, flowering and grain filling. Total performance index (PITOTAL), which describes potential of energy conservation within photosynthetic apparatus from exciton to the reduction of end acceptors of photosystem I, showed that barely cultivars differed in their response to drought stress. PITOTAL increased from booting to grain filling stage in most of the cultivars except for nine cultivars where decline was observed. In order to estimate the contribution of the different components (density of reaction centers, trapping to dissipation ratio and efficiency of excitation energy conversion to electron transport and performance index) to PITOTAL value, we transformed data to logarithmic values to obtain the parameter of partial driving forces. Stress conditions during growth season induced decrease of total driving force (DFTOT) in six cultivars at flowering stage. Dissipation and density of reaction centers mainly contributed to the lower value while efficiency of excitation energy conversion to electron transport and performance index mainly contributed to higher PITOTAL value in all other cultivars. At grain filling stage all cultivars, except one, revealed positive total driving forces. In conclusion, obtained results suggested that at flowering stage of growing, barley cultivars were more sensitive

to stressful conditions comparing to grain filling stage during which plants exhibited more stability in processes within photosynthetic apparatus indicating an adaptation to stress conditions.

**KEYWORDS:** barley, performance index, photosynthesis, development, drought stress

Poster presentation 36 01 39

### EFFECT OF DIFFERENT SELENIUM FORMS ON THE GLUTATHIONE METABOLISM IN WHEAT SEEDLINGS (*TRITICUM AESTIVUM* L.)

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Agronomic bio-fortification of wheat with selenium is a successful approach in improving its content in wheat grains. Due to its chemical similarity to sulphur, selenium is metabolized via sulphur assimilation pathways in plants. Therefore, in addition to oxidative stress induction, the application of selenium could also disturb protein structure and consequently influence glutathione metabolism in plants. Glutathione is involved in different processes such as biosynthetic pathways, detoxification of xenobiotics, antioxidant defence and maintenance of redox homeostasis. The aim of this study was to determine the impact of different concentrations of selenate and selenite on the glutathione metabolism in wheat seedlings (*Triticum aestivum* L.). For this purpose, contents of total, reduced and oxidised glutathione were determined, as well as the activities of the enzymes included in the glutathione metabolism such as glutathione reductase, glutathione peroxidase and glutathione S-transferase. Results showed that both forms of selenium affected total, reduced and oxidised glutathione content and activities of different enzymes in wheat seedlings. The glutathione content and enzyme activities depended on applied concentration of selenate and selenite. Both forms of selenium had more significant impact on the measured parameters in the root compared to wheat shoots, suggesting tissue-specific response. Obtained results can serve as a basis for further research on selenium toxicity and detoxification mechanisms in wheat. New insights into the selenium impact on glutathione metabolism could contribute to the further development of bio-fortification strategies.

**KEYWORDS:** glutathione, glutathione metabolism, selenium, wheat

Poster presentation 37 01 45

### THE EFFECT OF PHYTOHORMONES ON GROWTH AND BIOMASS PRODUCTION IN *HYPERICUM BARBATUM* JACQ. CALLUS CULTURES

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Phytohormones and their synthetic analogues represent the most important exogenous factors in regulation of proliferation and morphogenesis of plant cells in *in vitro* conditions. This is the first report for the effect of auxins 2,4-dichlorophenoxyacetic acid (2,4-D) and 1-naphthaleneacetic acid (NAA), as well cytokinin N6-benzyladenine (BA) on the growth and biomass production of *Hypericum barbatum* Jacq. callus cultures. For the establishment of *H. barbatum* callus cultures, epicotyls as primary explants were isolated from *in vitro* grown seedlings and inoculated on MS/B5 medium supplemented with different concentrations of auxins and cytokinins. In the experiment conducted, successful induction of calli (callogenesis) was observed from the wounded sites of epicotyls. Epicotyl-derived calli were isolated and monthly subcultured on the same nutrient medium used for callogenesis. After the third subculture, one-month old calli were used for determination of fresh biomass (FW) and dry biomass (DW) production, as well as growth index (GI). Obtained results showed significant differences in FW, DW and GI of calli cultured on different combinations of auxins and cytokinins. The calli cultured on medium supplemented with 1 mg·L<sup>-1</sup> 2,4-D and 0.1 mg·L<sup>-1</sup> BA, 1 mg·L<sup>-1</sup> NAA and 1 mg·L<sup>-1</sup> BA, as well 1 mg·L<sup>-1</sup> 2,4-D, 0.1 mg·L<sup>-1</sup> BA and 0.1 mg·L<sup>-1</sup> NAA exhibited the highest values for FW (2.2 g) and DW (0.18 g) compared to those grown on other tested combinations of phytohormones. Also, the GI value of calli grown on 1 mg·L<sup>-1</sup> NAA and 1 mg·L<sup>-1</sup> BA (12.8) was significantly increased in comparison to callus cultures grown on other tested media (from 3.6 to 8.3). The data indicated that exogenous application of high concentration of auxins (2,4-D and NAA) in combination with cytokinin BA is an effective approach for successful induction and proliferation of *H. barbatum* callus cultures. This study represents a solid base for further investigation of morphogenetic potential of *H. barbatum* for its *in vitro* culture establishment, for the purpose of biologically active secondary metabolites production.

**KEYWORDS:** Callus cultures, *Hypericum barbatum* Jacq., N6-benzyladenine, 1-naphthaleneacetic acid, 2,4-dichlorophenoxyacetic acid.

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### DIFFERENCES IN THE ANTIOXIDATIVE RESPONSE OF THREE SOYBEAN CULTIVARS TO CADMIUM

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Anthropogenic activities increased cadmium content in agriculture soils leading to its entry into the food chain, thus causing numerous pathological consequences for the consumers. In addition, cadmium causes changes in physiological and biochemical processes in plants affecting their growth and yield. Different plant species, but also different cultivars of the same species, vary widely in the terms of cadmium accumulation and tolerance. Considering it, production of soybean (*Glycine max* L.) as a leading oil and protein culture is endangered by low cadmium tolerance. Therefore, the aim of this study is to determine differences in the biochemical mechanisms of cadmium tolerance in leaves of three Croatian soybean cultivars (Korana, Ika and Lucija), which differ in the capacity of cadmium accumulation. As the indicators of oxidative stress, content of H<sub>2</sub>O<sub>2</sub> and lipid peroxidation levels expressed in terms of thiobarbituric acid reactive substances were determined, while antioxidative status was determined by the activities of antioxidative enzymes such as catalase, ascorbate peroxidase, glutathione peroxidase and guaiacol peroxidase. Cadmium treatment increased lipid peroxidation levels in leaves of all three soybean cultivars, indicating presence of oxidative stress. Also, the activity of glutathione peroxidase was increased in all cadmium treated plants, while other enzymes showed different activity patterns in investigated cultivars. Cultivar Korana, that showed the highest level of cadmium accumulation, also demonstrated the highest level of tolerance in regard to other two cultivars. The most sensitive cultivar Ika with moderate accumulation capacity showed the lowest level of antioxidative response. Biochemical mechanisms of tolerance to cadmium stress differed in three investigated soybean cultivars, and tolerance to cadmium was partly achieved by increased antioxidative response.

**KEYWORDS:** antioxidative response, cadmium, oxidative stress, soybean, cultivars

Poster presentation 39 01 13

### THE EFFECTS OF EXOGENOUS MOLYBDENUM APPLICATION ON NITROGEN METABOLISM IN WHEAT ROOTS EXPOSED TO CADMIUM STRESS

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Nitrogen is constituent of the enzymatic proteins which are essential for metabolism in seeds during germination and plant growth. Molybdenum (Mo) plays an essential role in molybdoenzymes in plants, such as nitrate reductase, aldehyde oxidase, xanthine oxidase and nitrogenase. Cadmium (Cd) is widely recognized as one of the most toxic heavy metals in the environment due to its high mobility. The important enzymes in nitrogen metabolism are nitrate reductase (NR), nitrite reductase (NiR), glutamate synthase (GOGAT) and glutamate dehydrogenase (GDH). We aimed to determine the effects of Mo on nitrogen metabolism (NR, NiR, GOGAT and GDH) of wheat roots exposed to Cd stress. Also, we analysed the growth, proline content and lipid peroxidation (TBARS content). Three-weeks-old wheat plants were treated with 5 and 10 μM Mo and with or without Cd application in growing medium (150 and 300 μM) for 7 days. The growth (RGR), proline content (Pro), the activities of nitrate reductase (NR), nitrite reductase (NiR), glutamate synthase (GOGAT) and glutamate dehydrogenase (GDH) and lipid peroxidation (TBARS content) were analysed. A decrease in the activities of NiR and GDH and lipid peroxidation were observed in response to increasing levels of Cd concentration. On the other hand, stress caused a reduction in RGR and Pro content. These changes were more pronounced in 300 μM Cd stress-treated wheat roots. However, in Cd-stressed wheat, Mo application resulted in alleviation on growth and TBARS content. Also, Mo application to Cd-stressed wheat roots significantly enhanced the activities of NR, GOGAT and GDH. It could be concluded that practice of exogenously applied Mo could be used for a reduction of heavy metal stress symptoms and improved growth and enzyme activities including nitrogen metabolisms in wheat roots.

**KEYWORDS:** cadmium stress, nitrate reductase, nitrogen metabolism, proline content, *Triticum aestivum*

### THE ROLE OF ROSMARINIC ACID IN PROTECTION OF OXIDATIVE DAMAGE INDUCED BY EXCESSIVE CHROMIUM IN MAIZE ROOTS

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Chromium (Cr) is one of the most toxic metals to plant metabolism causing photosynthesis and growth reduction when is accumulated in excessive concentrations. Organic acids are involved in detoxification mechanisms important for plant's physiological strategy in adaptability and tolerance to polluted environment. Rosmarinic acid (RA) is one of the water-soluble phenolic acids. The general aim of the present study was to understand the responses of applied RA to growth, osmolyte accumulation, some enzymatic and non-enzymatic antioxidants including ascorbate glutathione cycle and lipid peroxidation in Cr-treated maize roots. Germinated maize seedlings were grown under Cr stress (150 and 300  $\mu\text{M}$ ) in individual treatments and in combination with RA (50 and 100  $\mu\text{M}$ ; Cd+RA treatments). The growth rate (RGR), proline content (Pro), osmotic potential ( $\Psi_{\text{II}}$ ), enzymatic antioxidants (MDHAR and DHAR), non-enzymatic antioxidants, like ascorbate (tAsA), reduced and oxidized glutathione (GSH and GSSG), dehydroascorbate (DHA), and also, lipid peroxidation (TBARS content) were investigated. The results showed that Cd-stress caused a reduction in RGR,  $\Psi_{\text{II}}$  and Pro content. After exposure to Cr stress, the significant reductions of the MDHAR and DHAR and the contents of GSH and GSSG were observed in maize roots. Also, Cr excess in plant tissue caused an increase in the contents of DHA, tAsA and lipid peroxidation (TBARS). Under the increased rate of RA application, the oxidative stress induced by Cr was reduced, providing an increase in MDHAR, DHAR, the contents of GSH and DHA, and decrease in TBARS and the contents of GSSG and tAsA when compared to the stress alone. The obtained data indicated that addition of RA to growing medium can provide protection against the adverse effects of Cr stress by modulating growth, osmotic adjustment and ascorbate glutathione cycle in roots of young maize seedlings.

**KEYWORDS:** *Zea mays*, rosmarinic acid, chromium, proline, ascorbate-glutathione cycle, TBARS content.

### STRESS BIOMARKER: MICROTUBULE CYTOSKELETON RESPONSES OF THE SEAGRASSES *HALOPHILA STIPULACEA* AND *CYMODOCEA NODOSA* TO Ag NANOPARTICLE TOXICITY

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The toxic effects of silver nanoparticles (AgNPs), an ultimately fast-growing pollutant, were identified by assessing microtubule (MT) organization as a possible stress biomarker. The seagrasses *Halophila stipulacea* (Forsskål) Ascherson (1867) and *Cymodocea nodosa* (Ucria) Ascherson (1870) were used as experimental plant material. The experiments were conducted, under laboratory conditions, at concentrations starting from levels usually found in the marine environment to higher exposure levels (0.002, 0.02, 0.2  $\text{mg L}^{-1}$  AgNPs). Tubulin immunostaining was performed in meristematic and differentiating leaf cells of both seagrasses. All fluorescent specimens were examined with a Zeiss LSM780 CLSM and images were acquired with the ZEN2011 software. In addition, the characterization of AgNP size and structure and the effects on the seagrass cells were investigated by Transmission Electron Microscopy (TEM). In the untreated cells of *C. nodosa*, cortical MTs formed a dense network with diverse orientation, while many mitotic cells were observed. In *H. stipulacea*, cortical MTs exhibited uniform orientation, while fewer mitotic cells existed, compared to *C. nodosa*. The first toxic effects of AgNPs on *H. stipulacea* cells were detected on the 8th exposure day, only at the highest concentration (0.2  $\text{mg L}^{-1}$ ). MTs presented both a decline and a residual effect in the meristematic leaf zone, while they were completely depolymerized with a tendency to disappear in differentiating cells. The effects on *C. nodosa* cells were similar to those on *H. stipulacea* in both meristematic and differentiating cells, at the same exposure time, but at lower concentration level (0.02  $\text{mg L}^{-1}$ ) compared to *H. stipulacea* treatments. Concerning ultrastructural defects due to AgNPs, TEM observations revealed that *C. nodosa* chloroplasts were affected only at the highest concentration applied. In conclusion, the findings of this study suggest that MT organization and integrity is suitable as an early stress biomarker, while further targets of AgNP toxicity are revealed. This biomarker could be used for monitoring programmes for protection and management of coastal systems.

**KEYWORDS:** microtubules, seagrasses, stress biomarker, silver nanoparticles, toxicity, ultrastructure

### MOLYBDENUM ELIMINATES THE EFFECTS OF STRESS IN CADMIUM-TREATED WHEAT PLANTS THROUGH REGULATION OF WATER STATUS AND PHOTOSYNTHESIS

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Molybdenum (Mo), an essential micronutrient for plants, plays a role as electron donor/or acceptor in molybdoenzymes in plants, such as nitrate reductase, aldehyde oxidase, xanthine oxidase and nitrogenase. On the other hand, cadmium (Cd) is known to be a toxic metal that can cause severe damage to plants. However, little is known about the defensive mechanisms of Mo in plants under Cd stress conditions. This study was designed to investigate the effects of Mo on Cd-induced oxidative stress in wheat (*Triticum aestivum* L.) leaves. Plants were grown in nutrient solution containing Cd (150 and 300  $\mu\text{M}$ ) and/or Mo (0.15 and 0.3 mM) for 7 days (d). The water content (RWC), growth rate (RGR), proline content (Pro), osmotic potential ( $\Psi_{\text{II}}$ ), photosynthetic parameters (the maximum quantum yield in the dark-adapted state (Fv/Fm), the actual quantum yield in the light-adapted steady state ( $\Phi\text{PSII}$ ), the coefficients of photochemical quenching (qP) and the non-photochemical quenching values (NPQ)) and lipid peroxidation (TBARS content) were investigated in wheat leaves. The wheat leaves exposed to Cd stress exhibited a significant decline in RGR, RWC,  $\Psi_{\text{II}}$  and Fv/Fm,  $\Phi\text{PSII}$  and qP. The rate of this decline was more noticeable in treatment of 300  $\mu\text{M}$  Cd applied. Also, Cd stress treatment induced an increase in Pro content and NPQ. However, Mo application in wheat plants treated with Cd resulted in the alleviation of mentioned parameters in leaves. Compared with the plants grown in Cd stress conditions, exogenous Mo application markedly decreased TBARS content, and enhanced Pro content, RGR, RWC, Fv/Fm and  $\Phi\text{PSII}$ . Finally, our data confirm that Mo additional application to Cd-stressed plants has enabled plants to cope with stress-induced oxidative damage by protecting the growth, water usage status and photosynthetic apparatus.

**KEYWORDS:** cadmium stress, molybdenum, photosynthetic apparatus, *Triticum aestivum*, water content

### HUMIC ACID: REGULATION OF GROWTH, WATER STATUS AND NON-ENZYMATIC ANTIOXIDANT SYSTEM IN WHEAT LEAVES EXPOSED TO CADMIUM STRESS

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Humus compounds like humic acid (HA) produced in soil from organic residues have important role in stabilization of soil structure promoting at the same time plant development. The use of plant bio-stimulants based on the HA is an effective solution for elimination or mitigation of the oxidative damage derived from a cadmium (Cd) accumulation in plant tissues. The aim of this study was to assess the effects of HA (750 and 1500  $\text{mg L}^{-1}$ ) on growth (RGR), water content (RWC), osmotic potential ( $\Psi_{\text{II}}$ ), proline content (Pro), photosynthetic parameters (Fv/Fm,  $\Phi\text{PSII}$ , qP and NPQ), non-enzymatic antioxidants (ascorbate (tAsA), reduced and oxidized glutathione (GSH and GSSG), dehydroascorbate (DHA)) and lipid peroxidation (TBARS content) in HA-treated plants without Cd contamination and in HA-treated plants with Cd application at the same time (100 and 200  $\mu\text{M}$ ). Depending on its applied concentrations, Cd stress disrupted growth rate, water status, chlorophyll fluorescence and membrane structure in wheat leaves as indicated obtained results related to the excessive level of RGR,  $\Psi_{\text{II}}$ , Fv/Fm, Pro accumulation and TBARS content. Also, Cd stress caused decreasing in the contents of tAsA, DHA and GSSG, and NPQ levels during the experimental period. On the other hand, addition of HA to Cd-treated plants induced the increasing of values for RWC, RGR,  $\Phi\text{PSII}$ , qP and GSH content and GSH/GSSG ratio in wheat leaves. The increasing of these parameters by exogenously applied HA was in correlation with the reduction of TBARS. There was a positive correlation between HA application and the values of RGR,  $\Psi_{\text{II}}$  and Pro content, as well as with the parameters of antioxidant system. Thus, it could be concluded that HA application in contaminated soil by Cd increased the level of non-enzymatic antioxidant activity as well as the photosynthetic efficiency of wheat leaves.

**KEYWORDS:** ascorbate, heavy metals, humic acid, *Triticum aestivum* L., water content



### PHYTOTOXICITY OF ISOPROTURON TO NON-TARGETED AQUATIC PLANT *LEMNA MINOR* L. – A COMPARISON OF PULSED AND CONTINUOUS EXPOSURE

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Pesticides are usually repeatedly applied to agricultural surfaces and due to surface runoff and drainage may enter water environment. Therefore, pesticide contamination of water ecosystem mainly occurs in pulses, varying in concentration and duration. Standard toxicity tests used for essential risk assessment aim to maintain a constant concentration of tested substance through exposure period. On the other hand, tests that include experimental pulse scenarios alongside continuous exposure may provide information on latent herbicide effects and the potential of plants to recover or develop resistance to harmful herbicide effects. Therefore, the objective of the present study was to investigate the physiological changes in non-targeted aquatic plant *Lemna minor* L. caused by continuous and pulse isoproturon exposure. Isoproturon is one of the most frequently used phenylurea herbicides in conventional agriculture production, and it is often detected in surface water samples. To compare the effects of continuous exposure with pulse related effects different experimental designs were arranged. For continuous exposure treatment, a seven-day semi-static standard toxicity test was performed. Plants were treated with 0.05, 0.1, 0.15 and 0.2 mg L<sup>-1</sup> of isoproturon added to Steinberg's nutrient solution. For the pulse exposure treatment, plants were treated with equivalent isoproturon concentration for three days followed by four days of the recovery period. Herbicide concentrations for pulse exposure were calculated as time-weighted averages so that each pulse exposure scenario delivered a dose per week equivalent to that of the continuous exposure. Isoproturon caused a decrease in photosynthetic pigments content and protein concentration which in turn resulted in inhibited growth and significantly lowered growing and multiplication rate of plants. The growth rate of *L. minor* was considerably lower under continuous exposure compared with plants exposed to pulse herbicide treatment which may be due to a fast recovery in pulse exposure. Nevertheless, a concentration-dependent delay of growth rate in pulsed exposure was noted. Results from studies that include pulse exposure and recovery period may contribute to the further improvement of standard *Lemna*-test as a relevant form of environmental impact assessment.

**KEYWORDS:** isoproturon, *Lemna minor*, experimental pulse scenario, recovery, realistic risk assessment

### THE INFLUENCE OF SOME HEAVY METALS ACCUMULATION ON STEM ANATOMICAL CHARACTERISTICS OF *TRAPA NATANS* L. FROM SKADAR LAKE (MONTENEGRO)

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Metals are introduced into aquatic systems from natural and anthropogenic sources. Toxicity of some metals can be reduced by changing their chemical forms, but they forever remain in the environment. Contamination with metals is a widespread problem, which is also the case with Skadar Lake. The aim of the research presented in this paper was to analyze stem anatomical characteristics of *Trapa natans* from five Skadar Lake locations (L1 - Milovića bay, L2 - inflow of the Morača river, L3 - Kamenik, L4 - Grmožur, L5 - Lipovik) with different concentrations of Cu, Mn, Zn, Co and Pb. The middle part of internodes, from 10 plants, close to the leaf rosette were taken for anatomical analysis during the summer period of the year 2012. Cross sections of stem were made using cryotechnic procedure. Observations and measurements of stem parameters were performed using Carl Zeiss Imaging system Axio Vision Release 4.7. For all analyzed anatomical parameters, the minimum values at the location L2 were recorded, with the presence of maximum concentration for all investigated metals. On the other hand side, at the location L4 the minimum concentration for all investigated metals were recorded. Plants collected from this location have the largest average values of the most measured anatomical parameters. The results of Discriminant Analysis showed that plants from different location could be clearly classified into three groups according to their stem quantitative anatomical characteristics which corresponds with heavy metals concentration.

**KEYWORDS:** *Trapa natans*, stem, anatomy, heavy metals

### TRACE ELEMENTS PROFILES OF *NOCCAEA PRAECOX* AND *N. KOVATSI* GROWING ON ULTRAMAFIC AND NON-ULTRAMAFIC SUBSTRATES

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Multiple species of the genus *Noccaea* are well known metal-accumulators. However, the data on trace elements profiles within the species of this genus in the Balkans are scarce. The aim of this study was to analyze the ability of *N. praecox* and *N. kovatsii* accessions from ultramafic and non-ultramafic soils in the region of SE Europe to accumulate selected metals and to control their uptake and translocation. Concentrations of macronutrients (P, K, Mg, Ca and Fe) and eight trace elements (Ni, Zn, Mn, Cr, Co, Cu, Pb and Cd) were determined in the roots and shoots of analyzed plants, as well as in the rhizospheric soil from 20 sites. Biological concentration (BCF), accumulation (AF) and translocation (TF) factors were calculated to assess the accumulation potential of these species. All ultramafic accessions of *N. praecox* and *N. kovatsii* accumulated Ni in the shoots above the hyperaccumulation threshold set to 1000 mg kg<sup>-1</sup>, with the highest recorded Ni concentration in the sample of *N. praecox* from mountain Maljen in NW Serbia (14593 mg kg<sup>-1</sup> of dry plant weight). Nickel contents in all ultramafic accessions of both species were several fold higher in the shoots in comparison to the roots (TF >> 1), whereas values of the TF in non-ultramafic accessions were lower. Even though hyperaccumulation of Zn was not recorded in any of the examined samples, high values of AF and TF were calculated. The highest concentrations of Zn, exceeding 1000 mg kg<sup>-1</sup> dry weight were found in the shoots of *N. praecox* from mountain Čičarija (Istria, Croatia) and *N. kovatsii* from mountain Durmitor (Montenegro), both growing on non-ultramafic soils. Concentrations of Mn, Fe and Cr at all ultramafic sites were several fold lower in plant tissues than in the respective soils (AF << 1; BCF << 1). Clear differences in trace elements profiles among the samples of both species from different bedrocks could be explained by the physico-chemical properties of the soils, but could also be a result of adaptive traits related to specific edaphic conditions.

**KEYWORDS:** *Noccaea* species, SE Europe, trace elements, hyperaccumulation, accumulation factors

### ASSESSMENT OF HEAVY METAL POLLUTION OF TOPSOILS AND POTENTIAL ECOLOGICAL RISK IN CENTRAL URBAN ZONE OF BELGRADE

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The soil quality in urban and industrial zones is strongly negatively affected by human activities. Heavy metals in urban ecosystem mostly originate from fuel combustion and industry, and are consequently accumulated in the soil in different concentrations. In order to estimate the potential negative effect of heavy metals on the overall ecological balance in the city of Belgrade, analysis of heavy metal content (Ni, Zn, Cu, Co, Cd, Pb, Mn) in topsoils were performed. In total, seventy-two surface soil samples were collected from 18 sampling points in urban parks and in the proximity to central urban boulevards, during the four consecutive seasons in 2012. Concentrations of HM were determined using AAS. Measurements were taken in three replicates, and the obtained data were statistically analyzed using numerical descriptive analyses including mean (M) and standard deviation (SD). Pearson's correlation coefficient and Principal Component Analysis (PCA) was also performed. The obtained results indicate that the content of HM in soil samples vary amongst sites and seasons. The largest variations between sites are found for Pb, Cu and Zn, whereas moderate variations are found Ni, Co, Cd and Mn. The analysis show that concentrations of the investigated elements in soil samples taken from different sites within the same part of the city are directly negatively correlated to the distance from the traffic routes. In the surface soil layer there is a higher concentration of heavy metals, which are known as "urban metals", such as Pb, Zn, Cu and Cd. On the other side, there are also higher concentrations of metals that primarily have geological origin, such as Ni, Cr and Mn. The obtained preliminary results, as well as calculated ecological contamination factors, indicate that the index of potential ecological risk in Belgrade urban area is low.

**KEYWORDS:** heavy metals, topsoil, ecological risk

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### THE EFFECT OF APPLICATION OF HEAVY METALS AND N-ACETYL-L-CYSTEINE ON PHENOLIC CONTENT AND ANTIOXIDANT CAPACITY IN WHEAT SHOOTS

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Heavy metals (HMs) are a group of hazardous inorganic chemicals that cause contamination of agricultural land and soil. The most common HMs in polluted areas are lead (Pb), chromium (Cr), arsenic (As), zinc (Zn), cadmium (Cd), copper (Cu), mercury (Hg) and nickel (Ni). N-acetylcysteine (NAC) is derived from cysteine amino acid and has been used for many years as an antioxidant, free radical scavenger, chelating agent, and pharmaceutical drug, as well as nutritional support in treatment of diseases like metal poisoning and bronchitis. The aim of this study was to investigate the effect of treatment with different HMs and NAC on total phenolic compounds (TPC), phenolic acid (PA) content, and antioxidant capacity of wheat shoots. Wheat seeds were germinated in an incubator for one day, and seedlings were grown under hydroponic conditions at 25 °C and 70% humidity during the dark period. Five-day-old control and treatment group (NAC, Cu, Cd, Hg, Pb, Cu + NAC, Cd + NAC, Hg + NAC, and Pb + NAC) seedlings were harvested by using liquid nitrogen (-195 °C), and standard methods of analysis were used to determine TPC, free, ester, glycoside and ester-bound PA content, and antioxidant capacity (DPPH, FRAP and CUPRAC). TPC content in HM-stressed seedlings tended to increase in shoots in the form of: Hg>Cd>Pb>Cu compared to the control plants. The content decreased with NAC treatment compared to HM-stressed seedlings, except for Cd + NAC treatment. Antioxidant capacity values (DPPH, FRAP and CUPRAC) were correlated with TPC values. Increases and decreases in total PA contents varied depending on the type of HM. Comparing with the control, seedlings under the treatment with HMs promoted the PA synthesis, and this tendency of increasing PA was even more pronounced with addition of NAC (treatment: HMs + NAC), with some exceptions. Exposure of wheat seedlings to HM stress caused an increase in phenolic content (TPC and PA) and antioxidant capacity. NAC treatment caused different effects depending on HMs type.

**KEYWORDS:** wheat, heavy metal, N-Acetyl-L-Cysteine, phenolic acid, antioxidant

### BIOACCUMULATIVE AND TRANSLOCATION POTENTIAL OF THE SPECIES *POTENTILLA VISSIANII* (VIS. ET PANČ.) PANČ.

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*Potentilla visianii* (Vis. Et Panč.) Panč., the endemic plant of the central part of the Balkan Peninsula, grows on open steep rocky or lightly scattered black and white beech forests on the serpentinite geological background. The aim of this study was to determine the content of some elements (As, B, Cd, Co, Cr, Cu, Ni, Pb, Zn) in the soil and plant samples (root, stem, leaf, flower, whole plant). These elements are considered dangerous and harmful and may contribute to significant soil contamination. Determination of metal concentrations was performed by inductively coupled plasma mass spectrometry (ICP-MS). The obtained results indicate that the mean values of Cr and Ni concentrations in the tested soil are higher than the prescribed maximum permissible concentrations, limit values and remediation values, and the concentration of Co than the maximum permissible concentration and limit values for these metals in the soil according to the Regulation of the Republic of Serbia. The smallest content of almost all investigated elements (except Cd) was determined in the stem. The root contained the most of Zn, Ni, Cr, Co and Cd, while the leaf contained the most of As, B, Cu and Pb. The content of B was higher in all investigated organs compared to its content in the soil. The translocation factor greater than 1 was established at leaf for As, B, Cu and Pb, and at flower only for B. Also, it was shown that the leaves contained all the examined elements more than stem and flowers, while the flowers contained more investigated elements than stem (except Cd). The above-ground parts of the studied plant contained more As, B and Pb than the underground parts. The obtained results have shown that species *P. visianii* primarily accumulates Zn, Ni, Cr, Co and Cd in its roots, while As, B, Cu and Pb it translocates to leaves, and B in flowers, in which these elements are accumulated. The species *P. visianii* can be included in the list of potential candidates in soil remediation contaminated by As, B, Cu and Pb.

**KEYWORDS:** metals, *Potentilla visianii*, bioaccumulation, translocation

### GERANIUM ROBERTIANUM L. PLANTS TOLERATE HIGH LEVELS OF HEAVY METALS PRESENT IN THEIR NATURAL HABITATS

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Heavy metal toxicity causes several direct and indirect effects in plants such as growth retardation, inhibition of photosynthesis, DNA damage, alteration of secondary metabolism etc. Abiotic stress induces changes in plant metabolism by stimulating or suppressing the production of primary and secondary metabolites. Plants control heavy metal-induced stress by synthesis and accumulation of defense-related secondary metabolites, so the analysis of both secondary and primary metabolites content provides indication for understanding how plants manage stress. *Geranium robertianum* L. is quite an interesting object as it exhibits few limitations regarding the selection of soil. The objectives of the study were to explore *G. robertianum* ability to tolerate the presence of heavy metals in soil and to establish its potential as heavy metal hyper-accumulator. Plants were collected in flowering phase at four locations that differ in soil types (serpentine soil, anthropogenic soil, alluvial nitrogenated soil and soil rich in humus). Heavy metals concentration was determined from air-dried plant material by atomic absorption spectroscopy (Shimadzu 7000A) in acetylene/air flame (2200-2400° C). The contents of phenols, flavonoids, phenolic acids, proline and chlorophylls were detected by relevant methods based on spectrophotometry (UV-VIS Spectrophotometer; UV-mini 1240 (Shimadzu, Japan)). All experiments were performed in triplicate and expressed as mean values with standard deviation. Statistical analysis included analysis of variance and calculation of correlation coefficient (p<0.05). Plants collected from serpentine soil contained high levels of Fe, Zn and Mn with Ni concentration exceeding the level generally recognized as toxic to plants. Significantly increased level of secondary metabolites was detected in plants growing at nitrogenated alluvial soil while the highest concentration of chlorophylls was recorded in plants from anthropogenic soil. No significant correlation between heavy metals levels in plants and production of secondary metabolites and pigments was noticed. Proline levels were low in all tested plants indicating good tolerance of *G. robertianum* to heavy metal-induced stress.

**KEYWORDS:** *Geranium robertianum* L., heavy metal induced stress, secondary metabolites, chlorophylls

### BIOACCUMULATION CAPACITY OF *IRIS HUMILLIS*

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The mineral composition of the plant mainly depends on two factors – the growing soil composition and/or the influence of the environment (especially anthropogenic factor). Availability of metal's ions in soil for plant root is determined by element form, pH-value, etc. Some plants possessed ability to concentrate (bio accumulate) some macro- or microelements. The aim of this work was to examine ability of different parts of *Iris humillis* plant (rhizome, above-ground vegetative part and flower) to bound metal ions from surrounding growing soil or environment. This process was expressed as Biological Absorption Coefficient (BAC) which was calculated as  $c_{\text{plant part}}/c_{\text{soil}}$  ratio where  $c_{\text{plant part}}$  and  $c_{\text{soil}}$  are concentration of selected element in different parts of plant i.e. in soil. Content of selected elements (potassium, phosphorus and nickel) in soil and plant was determined by ICP-OES method (including pre-digestion of samples) and expressed as mg/kg of dry matter. Determined concentrations of potassium, phosphorus and nickel in soil were 877.1 mg/kg, 352.2 mg/kg and 2.97 mg/kg respectively. Content of selected elements in different parts of *I. humillis* was as follow: rhizome (K- 1686.0 mg/kg, P- 474.2 mg/kg and Ni- 8.5 mg/kg), above-ground vegetative parts (K- 3033.1 mg/kg, P- 423.1 mg/kg and Ni- 0.29 mg/kg) and flower (K- 1461.4 mg/kg, P- 273.0 mg/kg and Ni- 0.12 mg/kg). According to results for BAC potassium was bioaccumulate in all plant parts since value of this factor was higher than 1 ( $BAC_{\text{rhizome}} = 1.92$ ,  $BAC_{\text{above-ground vegetative parts}} = 3.46$ ,  $BAC_{\text{flower}} = 1.67$ ). Also, rhizome and above-ground vegetative part of *I. humillis* showed ability for bioaccumulation of phosphorus ( $BAC_{\text{rhizome}} = 1.35$ ,  $BAC_{\text{above-ground vegetative parts}} = 1.20$ ,  $BAC_{\text{flower}} = 0.78$ ) while rhizome possessed high ability to concentrate nickel ( $BAC_{\text{rhizome}} = 2.86$ ). The obtained results indicated that this *Iris* species possessed good ability for Ni-bonding through rhizome. It can be important for potential bioremediation use, since increased nickel concentration in the environment can be caused by some anthropogenic sources.

**KEYWORDS:** *Iris humillis*, bioaccumulation, potassium, phosphorus, nickel

Poster presentation 52 01 56

**THE EFFECTS OF YELLOW BEDSTRAW  
(*GALIUM VERUM* L.) WATER EXTRACTS ON  
SEED GERMINATION OF THREE WEED  
SPECIES**

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The widespread use of pesticides and their detrimental effects (soil pollution, pest resistance, etc.) have created a need for turning to natural resources in search for new/old methods of pest control. The inhibitory activity of water extracts of yellow bedstraw (*Galium verum* L.) flower, leaf, stem and root on seed germination of chickweed (*Stellaria media* (L.) Vill.), common ragweed (*Ambrosia artemisiifolia* L.) and velvetleaf (*Abutilon theophrasti* Medik.) seeds was examined. Yellow bedstraw plants were sampled near Krušedol Monastery, northern Serbia, on 7 July 2017, and then dried intact before the flowers, leaves, stems and roots were separated. Water extracts were prepared by extraction on a shaker for 24 h using 10 g plant material and 100 ml distilled water. Fifty seeds of each weed species were germinated in each of three replications in 10%, 20% and 50% extract solutions at their individual most adequate temperatures. The 20 and 50% concentrations of yellow bedstraw flower and leaf significantly reduced the final percentage of germinated chickweed, while the other two species were unaffected by any concentration. Stem extract had no decreasing effect on chickweed and velvetleaf germination, while the 10% concentration even had a significant stimulating effect on ragweed seed germination. Root extract concentrations of 20 and 50% reduced the final percentage of germinated chickweed seeds, while none of the tested concentrations showed effect on germination of the other two species. The extracts of all plant organs at each of the three concentrations slowed down chickweed germination speed. The flower and leaf extracts reduced common ragweed germination speed at 50% concentration, while velvetleaf germination speed was reduced by 20 and 50% concentrations. Stem extracts were ineffective regarding common ragweed and velvetleaf germination speed, and only the 50% root extract concentration affected on the speed of velvetleaf seed germination.

**ACKNOWLEDGMENTS:** The present study was part of projects TR 31043 and TR 31037 funded by the Ministry of Education, Science and Technological Development of the Republic of Serbia.

**KEYWORDS:** *Galium verum*, water extracts, weeds, germination, inhibition

**Session 2.**  
*Plant Taxonomy and Systematics*



Balkan  
Botanical  
Congress

Introduction lecture 01 02 22

## PLANT SYSTEMATICS AND SYSTEMATISTS – WHERE WE ARE, WHERE TO GO?

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Plant systematics is one of the oldest (but not old-fashioned!) botanical disciplines. Today, it is multidimensional and one of the most exciting and dynamic fields in biology. Systematics is not only significant for discovering, documentation and understanding of evolutionary history, but also because it provides the backbone of various biological and non-biological disciplines. Going back, it was believed that micro-morphic, evolutionary, biochemical, cytogenetic and most recently - molecular approaches, would resolve all phylogenetic and systematic issues. Did they? Certainly, the molecular approach has revolutionized systematics and many authors believe it is “The Holy Grail” in this field. However, some believe that relegating taxonomy, rich in broad knowledge and theory, to a high-tech services industry would be step backwards. Is “classical”, morphology-based or alpha-taxonomy, still relevant? Is taxonomy in crisis? What is the position of systematics in biology? Is it sufficiently in the education systems? With an ongoing planetary-scale biodiversity crisis, the need for systematists has never been greater. Systematists play an important role in the growing interest in biodiversity and conservation. Today, there is a lack of funding for taxonomic investigations. Generally, taxonomical studies have a low impact factor and, consequently, the number of taxonomists is permanently decreasing. However, botanists from the Balkans are enthusiastic even with insufficient funding. In Serbia, young scientists including systematists, are involved in scientific projects. Some key topics and challenges that emerged in recent years are: How to define a species? How many species exist on Earth? How they are related? What are the processes generating phytodiversity? With or without classical hierarchical categories? Which classification (phylogenetic) system to use? How should the next generation of systematists be trained? Plant systematics or Science of plant diversity? How to emphasize the prominent role of systematics to the outside world? Some recommendations for the future could be: recruitment of young scientists into systematics, collaborative work of systematists all over the world, establishing common standards for plant collecting and data sharing, herbaria digitalization, alpha e-taxonomy websites, struggle for permanent funding for long-term systematic studies, multi-disciplinary PhD and post-doc training, active communication with the general public, holistic approach to systematics.

**KEYWORDS:** plant systematics, systematists, biodiversity, future approaches

Introduction lecture 02 02 13

## DISENTANGLING RELATIONSHIPS IN DIFFERENT GROUPS OF *EUPHORBIA* SUBGEN. *ESULA* (EUPHORBIACEAE) FROM THE BALKAN PENINSULA

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*Euphorbia* is one of the largest genera of flowering plants including more than 2150 species. Most species in Europe belong to *Euphorbia* subgen. *Esula*, which is sister to three other subgenera. It comprises roughly 480 species and represents the most significant radiation of the genus in temperate areas of the Old World. One of the centres of diversity of this subgenus in Europe is the Balkan Peninsula, for which around 70 species have been reported, but the evolutionary origin of several of them remains poorly understood and their taxonomic status unresolved. We will present the outcomes of past and ongoing studies on evolution and diversification of several *Euphorbia* species from the Balkan Peninsula, based on DNA sequencing, AFLP fingerprinting, relative genome size measurements, chromosome number estimations and morphological analyses. More specifically, the diversification and evolution of the *Euphorbia barrelieri* group, *E. myrsinites*, *E. glabriflora* and *E. spinosa*, *E. niciana* and *E. seguieriana*, as well as *E. verrucosa* and *E. montenegrina* will be presented.

**KEYWORDS:** Balkan peninsula, *Euphorbia*, phylogenetic patterns, taxonomy

Introduction lecture 03 02 26

## PHYLOGEOGRAPHY OF THE AMFI-ADRIATIC *CAMPANULA GARGANICA* COMPLEX

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The Balkan Peninsula is one of the most important speciation and diversification centres in Europe. One of the morphologically and phylogenetically clearly recognized groups is the *Campanula garganica* complex. This complex exhibits amfi-Adriatic and Ionian distribution with the majority of species distributed along the western Balkan Peninsula. We here explore the phylogeography and diversification of the *C. garganica* complex based on nuclear (ITS), plastid (*trnL-trnF*) DNA sequences and AFLPs. The analysis of AFLP fingerprinting was performed on 39 populations in order to obtain further insights into phylogenetic relationships within the complex. In addition, phylogeographic data were complemented with habitat suitability models for present and last glacial maximum (LGM) period to infer the locations of putative LGM refugia. Phylogenetic study confirmed the monophyly of the complex, with poorly resolved phylogenetic relationship among taxa. The AFLP data clearly separated all investigated taxa revealing some new lineages, and indicated the existence of eight genetic groups. It also revealed hybridisation zones between *C. fenestrellata* and *C. istriaca* in the Northern Adriatic region, as well as the close relationship among *C. garganica* and *C. retina* from Italy, *C. cephalenica* and *C. acarnaica* from Greece while southern Adriatic populations of *C. teutana* and *C. poscharskyana* were connected to Albanian taxa *C. aureliana* and *C. skanderbegii*. The haplotype network detected 18 haplotypes and suggested that the taxa within the complex survived within different Balkan refugial areas which was consistent with the predictions of the LGM habitat suitability models. Constructed potential dispersal corridors during LGM between populations that share haplotypes indicated several highly isolated groups of populations with no apparent connectivity. The highest connectivity (both present and LGM) was identified between *C. fenestrellata* and *C. istriaca* populations in the Northern Adriatic and between *C. portenschlagiana* populations in the Central Adriatic. In summary, the obtained genetic structure of *C. garganica* complex supports connections between Balkan and Apennine Peninsulas probably during Pleistocene fluctuations of the sea level as well as the existence of areas in the Adriatic region.

**KEYWORDS:** *Campanula*, Balkan flora, LGM, evolution, phylogeography, systematics

Introduction lecture 04 02 56

## MORPHOLOGICAL VARIABILITY, EPIDERMAL STRUCTURES AND COMPOSITION OF EPICUTICULAR WAXES OF *SEDUM ALBUM* COMPLEX (CRASSULACEAE) IN BALKAN PENINSULA

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*Sedum album* is a succulent plant, with large phenotypic variability resulting in delimitation of a number of taxa with diverse taxonomic recognition. Variability as well as possibility of differentiation based on morphological, epidermal, micro-morphological and chemical characteristics of epicuticular wax of the species of “*Sedum album* complex” from Balkan Peninsula are considered. We analyzed the taxa from *Sedum album* in broad sense (*S. album*, *S. micranthum*, *S. athoum* and *S. serpentini*), to check whether the variability at different levels (morphology, epidermal structures, wax composition and micro-morphology) corresponds to their taxonomic reliability. The measurements of morphological characters were conducted using a dial caliper, stereo microscope and software Digimizer 8.0. Epidermal structures and epicuticular waxes were analyzed using light and scanning electron microscopy, gas chromatography with mass spectrometry and flame ionisation detector. We also used various multivariate statistical techniques in order to clarify the taxonomic status of identified taxa. Prior to the analysis, individuals of all collected species were subjected to a period of acclimatization in the greenhouse for at least one year. Morphological analyses appointed to moderate level of differentiation on the basis of upper leaf length, plant height, width of inflorescences, total number of flowers and nectaries width. Leaf epidermal studies of studied taxa have shown that significant systematic characters include adaxial epidermal and guard cell length, stomatal frequency and subsidiary cells area on abaxial epidermis. The distribution of *n*-alkanes C<sub>29</sub>, C<sub>30</sub>, C<sub>27</sub>, C<sub>32</sub> and C<sub>33</sub> was shown to be valuable criterion for their phytochemical delimitation. Results indicate presence of two (morphology and epidermal characters) or three (phytochemistry) well-defined groups of populations, with *S. serpentini* manifesting a considerable level of separation toward the others. *Sedum athoum* has shown weak to moderate degree of differentiation, while *S. micranthum* has shown general resemblance to *S. album*. The comparison of epicuticular wax microstructures has shown no significant differences. However, we believe that there are more characteristics (karyotype, molecular markers) to be considered in order to clarify taxonomic delimitation of the species from the complex from the Balkans.

**KEYWORDS:** *Sedum album*, variability, morphology, epidermal structures, epicuticular waxes, Balkans

Oral presentation 05 02 09

### A NEW SPECIES OF THE GENUS *TAXUS* (TAXACEAE) *T. TALYSCHENSIS* GUM., FROM TALYSH

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As is known, species of the genus *Taxus L.* have disjunctive area in various floristic regions. During the floristic studies in Talysh on the uppermost border of the forest, a centuries-old *Taxus* population of about 2 hectares was discovered. Species of the genus *Taxus*, which have survived to the present day, have not been sufficiently studied and there are no data on the composition of extinct species of this genus. Our palynological and morpho-anatomical studies of this kind show that the distinctive features between the described and recognized species of this genus are not too many. They do not even match for the species rank. The material for the study was based on owned collections, as well as collections of species of the genus *Taxus*, from the Herbarium of the Botanical Institute of the Russian Academy of Sciences, the Institute of Botany of Azerbaijan, as well as from other habitats. The anatomical and palynomorphological studies were carried out according to the generally accepted method. Different researchers use the fructification of the female and male organs, the pollen structure, morphological features and recently the molecular analysis as a diagnostic sign in the taxonomy of the *Taxus* genus and generally bare-grains. Unfortunately, the mentioned diagnostic factors do not fully prove themselves in these species of the plant taxonomy. The studies conducted throughout many years have shown that the stomatographic epithelial configuration (SKAN) and polynomorphological signs, as an unchanging diagnostic symptom, have a constant characteristic of the *Taxus* diagnosis. On the basis of all the diagnostic symptoms, a new type of *Taxus* species, *T. talyschensis* Gum., has been defined. The epithelial-cuticular, stomatologic, and palynological symptoms defined at the ultra-level are not identified in other types. The Talysh mountain system in the past geological era was not covered by glaciers, which allowed some representatives of tertiary vegetation to remain on Talysh. This species endemic to Talysh, has a narrow localization, which is typical of tertiary relicts.

**KEYWORDS:** Talysh, disjunctive, floristic, palynomorphological, stomatographic, rank

Oral presentation 06 02 01

### ACONITUM SUBSECTIO BURNATII SUBSECTIO NOVA

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A new subsection in the genus *Aconitum* includes 4 species in Europe, one of it is a Balkan endemic. *Aconitum* subsection *Burnatii* is presented here for the first time. The description of the new subsection and its scope is based on morphological and chorological investigations in all nature stands as on revisions of herbarium material. The blue flowering species of the genus *Aconitum* in Europe consist of 2 sections, *Aconitum* sectio *Aconitum* and sectio *Cammarum* (*A. degenii*, *A. lasiocarpum*, *A. pilipes*, *A. toxicum*, *A. variegatum*, *A. vitanum* etc.). Sectio *Aconitum* is separated into 2 subsections: *Aconitum* subsection *Aconitum* includes about 14 species in Europe (e.g. *A. anglicum*, *A. bucovinense*, *A. corsicum*, *A. firmum*, *A. napellus*, *A. plicatum*, *A. tauricum* etc.) and several species in Asia. *Aconitum* subsection *Burnatii* is endemic in Europe and consists of only 4 species, all of them well separated from each other in dispersed areas throughout Europe. *Aconitum burnatii* grows in the French Massif Central and in the southwestern Alps, *Aconitum maninense* is endemic in the Northern Carpathians (mainly in the area west and southwest of the Tatra mountains), *Aconitum nevadense* is a rare endemite of the Iberian peninsula (most of its only 20 populations are situated in the Sierra Nevada), and *Aconitum pentheri* is an endemite of the Balkans (Serbia, Montenegro, Makedonia, Albania and Bulgaria). There is one common characteristic for all 4 species, the straight glandular pubescence of the whole plant or at least of the inflorescence. Differences within these 4 species are the shape of the flowers, the shape of the nectaries, the shape of the leaves, the location of the glandular pubescence etc. Here all 4 species are defined by their morphological characteristics and are presented in their natural surroundings.

**KEYWORDS:** *Aconitum*, *Burnatii*, Europe, subsection, systematic

Oral presentation 07 02 14

### GENETIC DIVERSITY OF DIPLOID POPULATIONS OF *ARABIDOPSIS ARENOSA* AND *A. HALLERI* IN CENTRAL AND SE EUROPE

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Quaternary climatic oscillations had considerable impact on temperate biodiversity. For many species and areas, however, the consequences of this impact are still poorly known. In Europe, particular uncertainty surrounds the role of Balkans, a major hotspot of European diversity, in postglacial recolonization of more northerly areas, and the Carpathians, a putative northern 'cryptic' glacial refugium. Using combination of molecular methods (genome-wide SNPs, AFLP, nuclear microsatellites and sequences of single-copy nuclear regions and plastid DNA), we examined how the interplay of historical processes and niche shifts structured genetic diversity of diploid populations of *Arabidopsis arenosa* and *A. halleri*, members of the plant model genus that occupies a wide niche range from sea level to alpine peaks across considerable part of temperate Europe. While the northern Balkans hosted one isolated endemic lineage of *A. arenosa*, most of the genetic diversity of this species was concentrated further north in the Pannonian Basin and the Carpathians, where it likely survived the last glaciation in northern refugia. Finally, a distinct postglacial environment in northern Europe was colonized by populations of *A. arenosa* of admixed origin from the two Carpathian lineages. Niche differentiation along altitude-related bioclimatic gradients was the main trend in the phylogeny of *A. arenosa*. In the case of *A. halleri*, besides a distinct Alpine group, we identified a major phylogeographic split between Western and South-Eastern Carpathians, the latter group showing further genetic sub-structuring. Interestingly, the two Carpathian lineages were genetically closer to populations from neighboring mountain ranges (Sudeten + Hercynian mountains and Balkan mountains for the Western and South-Eastern Carpathian groups, respectively) than they were to each other, likely reflecting long-term isolation in different parts of the Carpathians and/or different (re)colonization pathways during the Holocene. Our study highlights the role of eastern central European mountains not only as refugia for unique temperate diversity but also sources for postglacial expansion into novel high-altitude and high-latitude niches. Knowledge of distinct genetic substructure of diploid *A. arenosa* also opens new opportunities for follow-up studies of this emerging model of evolutionary biology.

**KEYWORDS:** *Arabidopsis*, Brassicaceae, Cruciferae, postglacial colonization, glacial refugia

Oral presentation 08 02 15

### INTRODUCTION OF *CARDAMINE OCCULTA* INTO EUROPE AND ORIGIN OF THIS SPECIES

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The genus *Cardamine* is one of the largest genera of the Brassicaceae family, comprising about 200 species. Two species, namely *C. hirsuta* and *C. occulta*, are widely known as invasive ones, continuously spreading all over the world. Less known *C. occulta*, albeit currently one of the most widespread *Cardamine* species, had not until recently been recognised as a separate species. Schulz in his monograph treated populations of this taxon as subspecies of *C. flexuosa* With. (*C. flexuosa* subsp. *debilis* O.E. Schulz), but this was not followed by most of the subsequent authors. *C. flexuosa* was mostly treated as a single species without recognising any infraspecific taxa. In 2006, however, Lihová et al. showed that Eastern Asian weedy populations traditionally assigned to *C. flexuosa* represent a separate cytotype and genetic lineage. Recently, the name *C. occulta* Hornem. was determined for these Asian populations. While an octoploid level was ascertained for *C. occulta*, the tetraploid level was confirmed for *C. flexuosa* as well as their close relatives from Asia, *C. scutata* and recently described *C. kokaiensis* Yahara et al. The relative monoploid genome sizes were found to be strikingly different between these species. Variation in genome size agrees with different polyploid origins suggested based on genomic in situ hybridisation and comparative chromosome painting. These data indicate an autopolyploid origin of *C. kokaiensis* from a parental genome related to *C. parviflora* L. By contrast, an allopolyploid origin has been shown for the other species: *C. scutata* most likely originated via crossing of *C. amara* L. with *C. parviflora*, and *C. flexuosa* from *C. amara* and *C. hirsuta* L. Interestingly, *C. occulta* most probably originated through allopolyploid hybridisation of *C. scutata* and *C. kokaiensis*. While *C. occulta* is widespread in Asia and has been recorded in other continents for a long time, in Europe it was discovered only recently: in 1977 it was collected at Vercelli (Italy) and in 1993 in the province of Alicante (Spain). Nevertheless, currently it is present throughout most of the European countries, its spreading being supported mostly by horticulture. We are not aware of any record for *C. occulta* for the Balkan Peninsula, nevertheless, it is likely overlooked rather than absent in this area.

**KEYWORDS:** Brassicaceae, *Cardamine*, Cruciferae, invasive plants, polyploidy

Oral presentation 09 02 18

### ALYSSUM MONTANUM-A. REPENS GROUP (BRASSICACEAE) IN THE BALKANS, GENETIC AND MORPHOLOGICAL PATTERNS AND REVISED TAXONOMIC TREATMENT

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The high genetic, karyological and morphological complexity of the *Alyssum montanum*-*A. repens* species group (Brassicaceae) in the Balkans makes it a challenging study subject for exploring different speciation mechanisms and their taxonomic consequences. Ploidy level, genetic (AFLPs and chloroplast DNA sequences) and morphometric data were examined and confronted with recent taxonomic concepts. Remarkable genetic and morphological variation, which is often geographically structured, and high incidence of polyploids suggest a very complex evolutionary history in this area, involving allopatric differentiation and past hybridisation and polyploidisation events. A new taxonomic treatment, differing substantially from recent concepts, is suggested. Several Balkan taxa are clearly morphologically differentiated, namely *A. handelii*, *A. moellendorffianum*, *A. pirinicum*, *A. rostratum* and *A. wierzbickii*. Other species, *A. bosniacum*, *A. montenegrinum*, *A. reiseri* and *A. vernale*, exhibit overlaps in values of the most differentiating characters, but can all be steadily distinguished. *Alyssum repens*, which is not genetically compact, is still morphologically recognizable from other Balkan taxa. Two species, *A. austrodalmaticum* and *A. gmelinii*, despite having clear genetic differences and an allopatric distribution, only have minor morphological differences. Phylogenetic relationships among populations from the central Balkans and Greece are partly blurred apparently due to more extensive reticulations, and they are treated here under the tentative name *A. spruneri*.

**KEYWORDS:** *Alyssum*, Balkan Peninsula, Carpathians, molecular systematics, morphometrics, polyploidy

Oral presentation 10 02 27

### INSIGHTS INTO POLYPLOID EVOLUTION, RAPID RADIATION AND EXTENSIVE GENE FLOW IN DINARIC TAXA OF THE GENUS KNAUTIA (CAPRIFOLIACEAE)

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In recent years several phylogenetic and phylogeographic studies of different plant groups from southeastern Europe have revealed complex patterns in geographic distribution of genetic diversity, and have challenged traditional taxonomic concepts. *Knautia* section *Trichera*, which consists of mostly European perennial species with highest diversity in southern and southeastern Europe – especially the Alps and the Balkan Peninsula – presents a well-suited model to study evolutionary history and biogeographic patterns in the region. Recent research within the genus *Knautia* revealed a shallow genetic structure with mostly low resolution and only partial congruence between plastid and nuclear datasets possibly due to recent rapid diversification and ongoing hybridisation among different taxa. One of the revealed groups is an assemblage of several closely related taxa with distribution area restricted to Croatia and Bosnia and Herzegovina. The members of this group were previously assigned to the *K. dalmatica* and *K. velutina* groups. The *K. dalmatica* group is characterised by shortened internodes towards the stem base and regularly divided leaves with several linear to narrowly lanceolate lobes. It includes *K. adriatica*, *K. clementii*, *K. dalmatica*, *K. pectinata* and *K. travnicensis* from the Dinaric Mountains, all growing in (sub)mediterranean meadows and pastures. The *K. velutina* group is characterized by not or only weakly divided leaves with one to three pairs of lobes and a dense lanuginose to tomentose indumentum and – apart from the Apennine taxa – it includes *K. velebitica* from the Velebit Mts. All these taxa are morphologically very similar and difficult to differentiate. The aim of our study was to elucidate the relationships between the mentioned taxa as well as to answer the question, which taxa do really occur in the Dinaric Mountains. Ploidy-level screening of all investigated taxa revealed multiple cytotypes (di-, tetra- and/or hexaploids) within most of the taxa as well as similarity of genome sizes across different taxa belonging to the same ploidy level, with the exception of a few populations with divergent genome size. Preliminary morphological analyses confirmed the challenging differentiation based on leaf shapes and indumentum, while AFLP data revealed that most species do not form tight clusters.

**KEYWORDS:** *Knautia*, Dinaric mountains, AFLP, genome size, phylogeography

Oral presentation 11 02 58

### HOW COMPLICATED ACTUALLY IS GENUS ORNITHOGALUM?

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In most of the studies, genus *Ornithogalum* is characterized as „complicated“, and species are defined as conspecific. This is common in large sections such as sect. *Heliocharmos*, *Albuca*, *Dipcadi*, which have more than 30 species within them. The issue is evident if different identification keys are compared. Even species that have specific characteristics, i.e. *O. oligophyllum*, *O. refractum*, can easily be misidentified. In the same time, some investigations simplify genus diversity, aiming to describe most of infrageneric taxa as monophyletic. In that sense, from *Ornithogalum sensu lato*, 19 genera are recognized as monophyletic groups, with claim that they are easily recognized at morphological level as well. Other studies suggest that cytotype evolution went beyond phenotypic evolution. This is one of the explanations why cytotype diversity is above species diversity. All these „complications“ and inconsistencies in *Ornithogalum* taxonomy caused the need to change the problem focus. *Ornithogalum* s.l. is distinguished with simple morphology. In general, genus and its representatives can be described with few informative characteristics. Problem arises after 19<sup>th</sup> and 20<sup>th</sup> century, when more than 150 species were described only in sect. *Heliocharmos*. Based on that, in this study we try to find out which morphological characteristics are taxonomically important, and can be interpreted efficiently in identification key. We focused on sect. *Heliocharmos*, distributed in Mediterranean region, and far on the east, north and west of Asia and Europe. Material used for analysis is from Balkan Peninsula, Pannonia plain and neighboring regions. Overall, 26 species, 72 quantitative and 18 qualitative morphological and micromorphological characteristics are analyzed on fresh and pressed herbarium material. For analyses were used Multiple Correspondence Analysis, Principal Component Analysis, and Discriminant Function Analysis, all performed in Statistica ver. 13.3. Taxonomically important characters for morphological recognitions of sect. *Heliocharmos* and its species are presented, followed by combinations of characters that are useful for description of series or aggregates. In addition, characters that are too variable for quality interpretation, or cannot be recognized in dry specimens and in every phenophase are highlighted, as insufficiently descriptive. At the end, general identification key for sect. *Heliocharmos* is presented.

**KEYWORDS:** *Ornithogalum*, Hyacinthaceae, Ornithogaloideae, identification key

Oral presentation 12 02 17

### TAXONOMIC AND PHYLOGENETIC RELATIONS OF ALLIUM CHAMAESPATHEM BOISS., A REMARKABLE SPECIES OF BALKAN FLORA

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Based on recent systematic and phylogenetic researches, the genus *Allium* L. currently includes 920 species arranged in 15 subgenera and 15 sections. Among these groups, subgen. *Allium* is the largest one with over 300 species, 130 of which belong to the sect. *Allium*, well supported as monophyletic. Within this section, *Allium chamaespauthum* Boiss. stands out as the only autumn-flowering species, considered a Tertiary wild relative of a potential gene donor to many *Allium* crops, including *A. cepa*, *A. fistulosum*, *A. porrum*, *A. sativum* and *A. schoenoprasum*. It represents a typical Balkan element, occurring in several localities of Albania, mainland Greece, Peloponnese, Evvia, Crete, Ionian and Aegean islands, extending to South Italy, and growing in rocky places, usually garrigues and grasslands, rarely open woodlands and dunes. This species is very rare, with restricted populations. Literature data, herbaria examination, and new field surveys throughout its distribution area revealed a significant morphological variation among populations which suggested more detailed bio-systematic investigations. To clarify the taxonomic position of this peculiar *Allium*, and assess the variability range of its populations, living plants from several sites were analysed. Qualitative and quantitative morphological features were measured under stereomicroscope (6–66×) from both fresh and herbarium materials; karyotypes were obtained and characterized from somatic mitotic plates of root meristematic cells, through traditional karyotyping methods; leaf anatomy was studied on leaf cross sections from leaf blades of maximum size and optimal vegetative development; seed coat microsculpturing was assessed by SEM technique; for phylogenetic investigations, DNA extractions by CTAB2× and PCR amplifications of *nr*- and *cp*-DNA sequences using specific protocols were performed. Obtained sequences for ITS, *trnL-trnF* and *trnH-psbA* genes were assembled to generate MP, ML, and BI phylogenetic trees, and TCS haplotype networks. Though all populations share the same chromosome count  $2n=16$  and the typical karyotype structure of sect. *Allium*, with chromosomes having long linear satellites, relevant macro- and micro-morphological features, different ecological requirements, and significant genetic diversity, suggest that *A. chamaespauthum* should be treated as a complex species, with

several populations, from Crete, Albania, S Italy for instance, needing a deep revision of their taxonomic status.

**KEYWORDS:** *Allium chamaespathum*, Balkan flora, biosystematics, phylogenetics, section *Allium*

Oral presentation 13 02 39

### INTROGRESSION, HYBRID SPECIATION AND CROSSING BARRIERS IN CROCUS L. (IRIDIDACEAE)

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The genus *Crocus*, comprising about 200 species, requires a taxonomic revision as several infrageneric and even intraspecific units were shown to be para- or polyphyletic and over 50 new species were described recently but, in most cases, not safely assigned to existing taxonomical groups. Moreover, as up to now phylogenetic investigations failed to produce a resolved backbone phylogeny of the genus the affiliation of several species and infrageneric groups is still unclear and it is not possible to establish a meaningful new classification. To obtain such a phylogeny, 19 chloroplast and nuclear markers are currently sequenced using high-throughput technologies. Such multi-locus analyses increase the chance for resolved phylogenies and also allow the identification of hybridization events. Natural hybridization and hybrid speciation was previously assumed to be rare in *Crocus*. However, incongruences between different markers as well as the presence of two alleles, of which one is shared with another species, indicate that it might be much more common. In contrast to many other plant groups, where hybridization involves also allopolyploidization, in *Crocus* hybridizations seem to happen mostly on the homoploid level. However, for several sympatric *Crocus* species the existence of efficient crossing barriers is evident.

**KEYWORDS:** multi-locus phylogeny, hybrid speciation, *Crocus*

Oral presentation 14 02 38

### GEOMETRIC MORPHOMETRICS OF PERIGONE SEGMENTS SHAPE IN DIFFERENT SPECIES FROM SERIES VERNI MATHEW (CROCUS L. IRIDACEAE)

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Series *Verni* represents one of the taxonomically most complex series in the genus *Crocus*. It comprises six taxa: the *Crocus vernus* aggregate (split into five species – *Crocus vernus*, *C. neapolitanus*, *C. neglectus*, *C. siculus*, *C. heuffelianus*), *C. ilvensis*, *C. etruscus*, *C. kosaninii*, *C. tommasinianus* and *C. longiflorus*. This study included 160 specimens from 16 populations of five species from series *Verni* (*C. vernus*, *C. heuffelianus*, *C. neapolitanus*, *C. tommasinianus* and *C. kosaninii*) collected throughout the Balkan Peninsula. In order to assess the diversity of perigone segment shape, the landmark – based geometric morphometrics method was applied. To depict the shape of 160 outer and 160 inner perigone segments, 10 landmarks were selected – four bilaterally symmetric and two medial. The results of ANOVA and MANOVA revealed statistically significant differences between species. In the morphospace defined by the first two PC axes (95.91% for outer and 96.97% for inner perigone segments), two formations of species can be detected: *C. tommasinianus* and *C. kosaninii* with the wider, elliptical and rounded perigone segments, whereas *C. heuffelianus* and *C. vernus* are grouped together, having narrower, obovate perigone segments. With its central distribution, *C. neapolitanus* tends to be an intermediate between different morphotypes. Our investigation revealed not only the existence of good differential characters significant for identification of species, but provided also insights on the evolutionary history.

**KEYWORDS:** *Crocus*, *Verni*, perigone, geometric:morphometrics

Oral presentation 15 02 02

### TAXONOMY AND BIOGEOGRAPHY OF BRAMBLES (*RUBUS*, *ROSACEAE*) IN THE NW BALKANS

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The genus *Rubus* is one of the most complicated groups of vascular plants in Europe due to morphological and taxonomic complexity. The bramble flora of the NW Balkans remained nearly unexplored to date, thus, the main goals of the study were the taxonomic clarification of the biotypes occurring in area, and the unravelling of evolutionary processes of key groups. During field excursions and herbarium studies several new species, among them those of new to the science, were found. Beside morphology-based taxonomic studies, the ploidy of all species was investigated, and certain groups were analyzed using molecular methods. The richness of ser. *Discolores* is especially high, both main (triploid and tetraploid) lineages play a key role in the bramble flora of the area. Significant unexpected hotspots for ser. *Micantes*, ser. *Sylvatici* and ser. *Vestiti* were demonstrated, some species of Balkan origin expand into the neighboring Pannonian and central European regions. The ser. *Rubus* is poorly represented in the area, mainly restricted to the northernmost mountains. In the especially intricate ser. *Glandulosi*, only putatively sexual biotypes are known, but they actively take part in hybridization processes mainly with members of ser. *Discolores*. Some sub-Atlantic groups (e.g. ser. *Pallidi* and ser. *Radula*) are completely missing or solely represented by local hybridogenous biotypes. Accepted species from the complete sect. *Corylifolii* (which are quite common in central Europe and still dominant in the Pannonian region) are restricted to the NW edge of the area – however, local hybrids without taxonomic value are quite common. Generally, the highest species richness, and the (mono)dominance of the apomicts, were observed in the mountains having broader belt of beech and oak-hornbeam forests; the flora of drier Mediterranean regions is reduced to few sexual species (*R. caesius* and *R. ulmifolius* agg.). The narrow intermediate zones or “rendezvous lines” (e.g. Istria, Papuk and Fruška Gora Mts.), where more climatic effects are in touch, are ideal regions for further studies due to formation of new taxa and the mixture of sub-Atlantic and Mediterranean floras. In some locations here, the number of local biotypes is much higher than those of the accepted species.

**KEYWORDS:** species creation, hybridization, apomixis, climatic barriers, colonization

Oral presentation 16 02 55

### RUBUS SUBGEN. RUBUS ON FRUŠKA GORA Mt. (SERBIA)

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Fruška gora is an isolated mountain on the southern edge of the Pannonian basin. It is on the meeting point of Pannonian and Continental biogeographic regions with some Illyrian sub-Mediterranean elements. These diverse influences gave rise to rich vegetation and high diversity of vascular plants. *Rubus* subgen. *Rubus* (brambles, blackberries) includes common European species with complicated taxonomy, history of ongoing hybridization and high variability of reproduction modes. Considering scarce knowledge of the *Rubus* subgen. *Rubus* in Serbia, and the Balkans in general, we aim to elucidate diversity, evolution and reproduction modes in brambles of Fruška gora. Field research was carried during 2016-2017. In addition to morphological analyses, flow cytometric seed screen was conducted to determine reproduction mode and Sanger sequencing of two non-coding plastid regions was used to reveal phylogenetic patterns. Bramble flora of Fruška gora is comprised of few basic taxa, several hybridogenous taxa with regional distribution and many local hybrids without taxonomical value. Out of around 20 morphotypes present, basic taxa are represented by widespread species mostly tolerating semiarid climate, such as *R. praecox*, *R. canescens* and *R. caesius*, as well as sub-Atlantic central European species (e.g. *R. montanus* s.l.). Second most common constituents of bramble flora are undescribed regional taxa from series *Micantes*, *Rhamnifolii* and *Discolores*. Remaining taxa are most numerous yet most infrequent. Some of these local hybrids were recorded only once as an individual bush (e.g. representatives from series *Vestiti* and *Pallidi*). Largest patches of bramble thickets are formed in half-shaded places, along forest roads, fringes and clearings, while only few are present in open ruderal habitats or under closed forest canopy. The most common ploidy level was tetraploid, followed by triploid (*R. ser. Discolores*) and only one diploid species. Diploid species *R. canescens* is exclusively sexual, whereas triploid species reproduce only by apomixis. Tetraploid representatives exhibit highest variability of reproduction modes, ranging from strictly sexual to facultative pseudogamous apomixis. Plastid haplotype diversity fits into general pattern observed in Central European brambles, pointing to common origin of the bramble flora on Fruška gora.

**KEYWORDS:** apomixis, brambles, reproduction, flow cytometry, Serbia, *Rubus*

Oral presentation 17 02 52

## PHYLOGENY OF VALERIANELLA (VALERIANACEAE)

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*Valerianella* Miller is a cosmopolite annual herbaceous genus with approximately 50 species distributed throughout America, Europe and Asia, with the centre of diversity in Anatolia and Russia. The genus is represented with 32 species - 2 of them are endemics - in Turkey, 35 species in Russia (25 of them are in Turkey also), 24 species in Iran (20 of them are in Turkey also), 22 species in Europe (17 of them are in Turkey also), and 8 species in America. Previous phylogenetic studies based on DNA sequences did not clarified infrageneric phylogenetic relationships of the genus because of limited sampling. There are only 17 species' molecular data in genbank. Additionally, there are controversies whether genus *Dufresnia* is synonym of *Valerianella* or not. To improve our knowledge of intrageneric and infrageneric relationships, here we present molecular phylogenetic analysis of *Valerianella* based on internal transcribed spacer (ITS) and *trnK* intron markers. In this study we sampled 48 additional accessions from *Valerianella* belonging to 26 species. Eighteen species of them are additional species accessions and we used more than 200 sequences of Valerianaceae from genbank. We conducted bayesian and maximum likelihood analysis by Beast and RAxML software respectively. We also discussed the floral and mericarp characters and their implications to our phylogenetic hypothesis. As a result, *Valerianella* is proved to be well supported as a monophyletic genus according to ITS dataset. In agreement with previous hypotheses *Dufresnia* is nested in *Valerianella*. One of our results is unexpected that some of American *Valerianella* species are nested in genus *Centranthus* according to *trnK* dataset. Furthermore, there are strong hints that hybridization and introgression have played a significant role in the evolution of the genus members in concordance with literature. We discussed deep insights into the infrageneric relationships, the correlation among species, morphological and geographical distributions.

**KEYWORDS:** *Valerianella*, phylogeny, ITS, *trnK*

Oral presentation 18 02 05

## MICROMORPHOLOGICAL AND CARPOLOGICAL INVESTIGATIONS ON SOME REPRESENTATIVE CARDUEAE (ASTERACEAE) TAXA FROM NE ANATOLIA

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The tribe Cardueae is represented by approximately 100 taxa indicated in 22 genera in the North East Anatolia. In this study, micromorphologies of fruit including pappus and anatomical characteristics of six taxa, belonging to five genera in the tribe *Cardueae* were investigated, in detail and all data are presented here for the first time. Plant materials used in this research were collected from natural habitats during the vegetation periods of taxa between 2011 and 2017. Morphological studies were carried out from herbarium materials. Mature fruits and pappus obtained from herbarium samples or randomly collected in fields were used in SEM analyses. Cypselas shapes of the investigated taxa are oblong, obovoid, oblanceolate and obpyramidal, and while the smallest cypselas was found in *Callicephalus nitens*, the biggest one was observed in *Centaurea helenioides*, and the widest fruit was detected in *Ptilostemon afer* subsp. *eburneus* was among the investigated taxa. Carpoidium was detected in the fruits of *Callicephalus nitens* differently from other related taxa. Based on cypselas surface ornamentation, scalariform, scalariform-reticulate, reticulate-tuberculate and rugulose fruits were determined. Pappus including caducous taxa, was obtained and their characteristics were given by using SEM analysis. It was observed that *Callicephalus nitens* has the biggest pappus and *Jurinea alpigena* has the smallest one. Pappus is determined as scabrous, subplumose and plumose, in addition, their bristles/hairs are present in above. On the other hand, it was determined that pappus diameter and frequency of bristles/hairs significantly differ among the investigated taxa. Pericarp anatomical structures were described, as well as the structure of testa. Sclerenchymatic pericarp presents in *Centaurea helenioides* and two *Psephellus* taxa, while other relatives have parenchymatic ones. Tetragonal crystals were observed in the pericarp of *Ptilostemon afer* subsp. *eburneus* and *Psephellus simplicicaulis*. Distribution of sclerenchymatic zone of the ribs and secretory ducts in mesocarp were evaluated. Data obtained from this study were compared the data present in literature. These results demonstrate that the compared fruit anatomical and micromorphological characters among taxa are significantly different and can be used as taxonomic markers in their classifications.

**KEYWORDS:** *Cardueae*, cypselas, anatomy, micromorphology, SEM, NE Anatolia.

Poster presentation 19 02 07

## GREEK ELEMENTS IN THE NOMENCLATURE OF THE FAMILY ASTERACEAE

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The name of the family Asteraceae comes from the Greek word "ἀστήρ", which means star. This example shows just a glimpse of the contribution of the Greek language to the scientific nomenclature. In order to identify this contribution to plant taxonomy, the taxa of family Asteraceae from the Greek flora were chosen aiming to study the Greek elements in the names of the genera, species and subspecies of the family. Asteraceae was selected because it is the largest family of the Greek flora, in terms of species richness, and it accounts for 13% of all Greek species. The genera, species, and subspecies of the family (1100 taxa) were retracted from the Vascular Flora of Greece; each and every name was checked for its possible Greek origin, using specialized dictionaries. All the Greek words were analyzed for their meaning and any possible connection with the biology of the taxon. Our preliminary results show that 67.3%, 37.9% and 42.9% of names of the genera, species and subspecies, respectively, are of Greek origin. The rest of the names come mostly from Latin words, whereas very few names have some other origin, like for example Anglo-Saxon. It is noteworthy that a quite large amount of names come from the surnames of acclaimed botanists: in Asteraceae genera 1.8% of the names come from the names of botanists, whereas this rate rises to 8.7% and 8.3%, to the species and subspecies, respectively.

**KEYWORDS:** Greek flora, Asteraceae, Greek elements, scientific nomenclature, plant taxonomy

Poster presentation 20 02 43

## REVIEW OF NOMENCLATIONAL NOTES OF THE NAMES RELATED TO *SILENE SAXIFRAGA* GROUP (CARYOPHYLLACEAE)

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Great morphological variability of the *Silene saxifraga* group goes with complex taxonomy and nomenclatural issues re-

garding a high number of published names. Original descriptions in protologues were studied, as well as all other available literature sources where the names of the taxa belonging to this group were typified, in order to provide an overview of typifications and nomenclatural notes of the names related to this group. The following names are discussed herein: *Cucubalus alpestris*, *Saponaria caespitosa*, *Silene conglomeratica*, *Silene dionysii*, *Silene fruticulosa*, *Silene hayekiana*, *Silene notarisii*, *Silene multicaulis* var. *ottomanica*, *Silene oreades*, *Silene orphanidis*, *Silene paeoniensis*, *Silene parnassica*, *Silene parnassica* subsp. *vourinensis*, *Silene petraea*, *Silene pindicola*, *Silene saxifraga*, *Silene saxifraga* subsp. *balcanica*, *Silene saxifraga* var. *triflora*, *Silene serbica*, *Silene schmuckeri*, *Silene taygetea* and *Silene velcevii*.

**KEYWORDS:** nomenclature, original material, *Silene saxifraga* group

Poster presentation 21 02 08

## MORPHOLOGICAL VARIABILITY OF LEAVES OF *RIBES PETRAEUM* (GROSSULARIACEAE) IN EUROPE

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The occurrence of *Ribes petraeum* is limited to European mountain ranges, where it grows in dispersed localities. Depending on the taxonomic approach the taxon is sometimes divided into varieties or subspecies; to some of them the rank of independent species is often assigned (e.g. *Ribes caucasicum*). The aim of the present study was to biometrically verify differences in leaf characteristics between these taxa within its whole European range. The material came from: Pyrenees, Alps, Dinarides, Sudetes, Carpathians, and Balkan Peninsula. From each of the localities, over 30 leaves were randomly collected from short- and long-shoots. All the measurements were performed for the abaxial leaf side and included the following traits: petiole, midrib, middle lobe length; blade width; middle lobe width at its widest part and at the base; vein number of the middle lobe; lateral vein length; angle between midrib and lateral vein; basal angle. The standard deviation and coefficient of variation were calculated for each of the examined traits. The characteristics of intra-population variability were presented based on Discriminant Correspondence Analysis and Cluster Analysis. The analysis showed that the most variable are petiole length, midrib length, blade width, and basal angle; while the angle between midrib and lateral veins is the least variable. Statistical analysis facilitated differentiation of distinct geographical groups: Pyrenean, Alpine and Carpathian. Each of them is characterized by a specific combination of morphological traits. The differentiating traits are the length of petiole, blade dimensions, the length of lat-



eral veins, venation density, middle lobe shape. The research confirms the existence of three separate taxonomic units in the rank of varieties/subspecies: *atlanticum* (Pyrenees), *alpinum* (typical species for Alps and parts of Sudetes and Dinarides) and *carpathicum* (Dinarides, Carpathians, Balkans). Geographical isolation of mountain massifs and diversity of habitats and phytocoenoses probably led to the development of various morphological types. For example, “*carpathicum*” specimens with the largest leaves occur mainly in the Western Carpathians on humid and rocky habitats in sycamore forests. On the other hand, “*alpinum*” specimens, with distinctly smaller leaves, are present in the Alps and part of Dinarides, where they grow in subalpine shrubs.

**KEYWORDS:** rock currant, mountains, biometrics, morphology, systematics

Poster presentation 22 02 28

### MORPHOLOGICAL VARIABILITY OF BOLBOSCHOENUS GLAUCUS (LAM.) S.G. SMITH (CYPERACEAE) POPULATIONS FROM SERBIA

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The genus *Bolboschoenus* (*Scirpus*) is represented in Serbia with four species: *Bolboschoenus maritimus*, *B. glaucus*, *B. planiculmis* and *B. laticarpus*. *B. glaucus* is a freshwater thermophilic species that may be found along banks of large rivers. For a long time this species was included within *B. maritimus* s. l., under various names: *Scirpus macrostachys*, *S. tuberosus*, *S. maritimus* var. *glaucus*, *S. maritimus* var. *macrostachys*, *S. maritimus* var. *amentiferus*, *S. maritimus* var. *theta*, *S. maritimus* var. *terrestris*. Some previous studies on populations of *B. glaucus* noted high morphological variability in morphological characters of inflorescences but quantification of that variability and statistical comparison were not performed before this study. The aim of this study was to analyze and compare morphological variation among 12 populations of *B. glaucus* from Serbia (16 characters x 251 individuals). The standard univariate (Descriptive statistics) and multivariate statistics (PCA, CDA) were used for these purposes. Analyses have shown that apart from the overall morphological variability there were four morphologically distinct groups of populations. We assigned informal names to 4 groups of *B. glaucus* populations: *B. glaucus* “*macrostachys*”, *B. glaucus* “*astericum*”, *B. glaucus* “*intermedium*” and *B. glaucus* “*robustus*”. The most differentiated characters in these groups were: length of sessile spikelets, number of rays, number of ray spikelets and length of ray spikelets. These four characters have shown significant contrast between members of each pair of population

groups. The results of PCA analyses have shown that population of *B. glaucus* “*robustus*” was well-differentiated from all other populations and located in the negative part of first PCA axis and the positive part of the second PCA axis. *B. glaucus* “*intermedium*”, *B. glaucus* “*astericum*” and *B. glaucus* “*macrostachys*” were more closely associated in the center of ordination plot and there was a slight overlap between members of each pair. The CDA analysis has shown that populations of *B. glaucus* “*astericum*” and *B. glaucus* “*macrostachys*” represent two completely separate groups. Populations of *B. glaucus* “*intermedium*” and *B. glaucus* “*robustus*” have shown transitional characteristics between *B. glaucus* “*astericum*” and *B. glaucus* “*macrostachys*”. Further research is necessary for resolving the nature of morphological variability of *B. glaucus*.

**KEYWORDS:** *Bolboschoenus glaucus*, morphological characters, differentiation

Poster presentation 23 02 57

### MORPHOLOGY AND MICROMORPHOLOGY OF HELIANTHEMUM MILL. (CISTACEAE) SEEDS ON THE BALKAN PENINSULA

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*Helianthemum*, the largest genus of the Cistaceae family, is distributed in North Africa, Europe and Asia with the diversity center in the Mediterranean region. This genus is taxonomically complex since the main diagnostic characters (habitus, presence and shape of stipules, indumentum type, leaf shape and size, sepal size, petal size and colour) may vary both within and between populations due to phenotypic plasticity and local adaptations as well as introgression and hybridization. In the present study, seeds of seven *Helianthemum* taxa, collected on the Balkan Peninsula, belonging to both subgenera (*Helianthemum* and *Plectolobum* Willk.) were examined using light and scanning electron microscopy (SEM). Altogether six characters: three quantitative (seed length, seed width and ratio length/width) and three qualitative (shape, colour and testa structure) were analyzed. To test differences between quantitative characters, non-parametric Kruskal-Wallis test followed by Mann – Whitney pairwise comparisons were performed using the program Past ver. 3.11. The aims of this research were to study seed micromorphology of *H. nummularium* (L.) Mill., *H. oelandicum* (L.) DC. and *H. marmoreum* Stevan, Matevski & Kit Tan and to assess possibility of using seed characters for taxa delimitation between subgenera, species and subspecies in *Helianthemum* genus. Based on our observations, analyzed seeds could be separate in two groups by seed shape and testa structure: 1) ovoid, dense tuberculate, dull (dominant in *H. nummularium*, subg. *Helianthemum*) and 2) ovoid-triangular, minute tuberculate or pusticulate,

lustrous (dominant in *H. oelandicum* and *H. marmoreum*, subg. *Plectolobum*). Colours of the seeds covered spectrum of yellow, brown and dark reddish brown. The mean values revealed that *H. nummularium* has the largest seeds (1.72 x 1.36 mm) while smaller seeds were found in *H. oelandicum* (1.47 x 1.02 mm) and *H. marmoreum* (1.45 x 0.96 mm). Differences in the quantitative characters were statistically significant between two subgenera, while *H. nummularum* were the most different among species. Additionally, considerable differences were revealed in seed shape and testa structure which could be useful in distinguishing subgenera in *Helianthemum*.

**KEYWORDS:** light microscopy, *Plectolobum*, SEM, taxonomy, testa.

Poster presentation 24 02 53

### MORPHOMETRIC STUDIES ON SIDERITIS SUGGEST EXISTENCE OF A NEW SPECIES IN BULGARIA

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Section *Empedoclia* of genus *Sideritis* is a very complex taxonomic group with high percentage of endemism that have attracted the attention and provoked controversies among the scientists concerning the species delineation and identification. *S. scardica* is endemic for the Balkan Peninsula while *S. syriaca sensu lato* is believed to have wider distribution. Both species occur naturally exclusively on limestone, although can be cultivated on a broader range of soil pH. We present results of a study on morphometric variation in eight natural populations of *Sideritis* sect. *Empedoclia* occurring in Bulgaria. Thirteen measured traits and four ratios were used to reveal the degree and distribution of variation. Cluster analysis and Principal component analysis revealed that the two taxa, *S. scardica* and *S. syriaca* are well distinguished but the population Chervenata stena, classified as *S. scardica* differed significantly from all remaining populations in a complex of traits, the most differentiating being the ratios length:width of the first three uppermost leaf pairs. The raised progeny provided new evidence about the differences among the populations, especially in the hair density on the stem and leaf shape. Based on the results we consider that the population of Chervenata stena represents a different, new species.

**ACKNOWLEDGEMENT:** The authors express their thanks for the support provided by the project DFNP 17-17, Program for Career Development of Young Scientists, Bulgarian Academy of Sciences.

**KEYWORDS:** medicinal plants, phenotypic variation, taxonomy

Poster presentation 25 02 29

### MORPHOLOGICAL STUDIES ON CHAMAECYTISUS LASIOSEMIUS (BOISS.) PIFKÓ AND OTHER BALKAN CHAMAECYTISUS SPECIES

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*Chamaecytisus lasiosemius* described from middle north Turkey with some additional records from the Balkan Peninsula has been an insufficiently clarified taxon. It was missing from *Flora Bulgarica* and *Flora of Europe*, and it was synonymized with *Cytisus supinus* in *Flora of Turkey*. Cristofolini also accepted this species but he considered *Ch. frivaldszkyanus* as a synonym of *Ch. lasiosemius*. The purpose of the morphological study was to clarify the most important morphological characters of *Ch. lasiosemius*, which can clearly differentiate it from other taxa. *Chamaecytisus* specimens, including type specimens of *Ch. lasiosemius* (P 02952886) and other important species from the Balkan Peninsula, have been revised in the following herbaria: BP, PR, PRC, SO, SOM, W, WU. *Ch. lasiosemius* has branches and petioles with patent hairs, it is blooming in summer with flowers in terminal heads, its corolla yellow, vexillum densely hairy, calyx more or less sparse patent hairy, legume lanate, leaflets generally ovate, upper surface slightly hairy with short hairs. *Ch. lasiosemius* resembles *Ch. supinus* subsp. *supinus*, since branches and petioles have same patent hairs. However, vexillum of *Ch. supinus* is generally glabrous, rarely slightly hairy, but it is densely hairy in *Ch. lasiosemius*. Some *Chamaecytisus* species distributed in the Balkan Peninsula have flowers in terminal heads, branches with patent hairs and their vexillum is densely hairy. According to the hairs on branches these species can be grouped as follows: 1. The *Ch. rochelii* group (*Ch. virescens*, *Ch. kovacevi*, *Ch. danubialis*), which has straight hairs of various length, covering the young branches densely. 2. The *Ch. frivaldszkyanus* group (*Ch. calcareus*), which has appressed and patent hairs, covering the young branches also densely. Some of the patent hairs are parallel with the branches, but these hairs are broken at a blunt angle. 3. The *Ch. lasiosemius* group, which has more or less frizzy hairs, not covering the branches densely. Based on our morphological study we can conclude that *Ch. lasiosemius* clearly differs from all other *Chamaecytisus* taxa, thus we accept it as a separate species with a distributional area of North Albania, Bulgaria and Turkey.

**KEYWORDS:** Albania, Bulgaria, *Cytisus*, Fabaceae, taxonomy, Turkey

Poster presentation 26 02 42

### MORPHO-ANATOMICAL DIFFERENTIATION OF *TEUCRIUM MONTANUM* L. (LAMIACEAE) IN THE BALKAN PENINSULA

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*Teucrium montanum* L. is evergreen microphyllic xeromorphic dwarf shrub. It is distributed in the Balkan Peninsula, extending from the sea coast to altitudes of over 2.100 m a. s. l. with the highest number of occurrences in the zone between 500 and 1.000 m. Due to its high morphological variability, several species have been described in the Balkan Peninsula (*T. helianthemoides* Adamović, *T. pannonicum* A. Kerner, *T. parnassicum* (Čelak.) Wettst., *T. skorpilii* Velen., *T. supinum* L. etc.). However, the taxonomic status of these taxa is not clear, and in modern floristic literature and checklists they are considered as synonyms or infraspecific taxa within broadly defined *T. montanum*. Taking into account the unresolved taxonomic relationships among Balkan populations of *T. montanum* the aim of this study was to investigate the morpho-anatomical variability and differentiation of 13 populations from Serbia (SR-Trešnjica canyon, SR-Brdjanska gorge, SR-Goč, SR-Gornjak gorge, SR-Grza canyon, SR-Jelašnica, SR-Kopaonik, SR-Maglič, SR-Rtanj), Greece (GR-Olimp, GR-Ossa) and Albania (AL-Deja, AL-Skadar). Therefore, we analyzed 23 characters of leaf anatomy and 16 characters of stem, leaf and calyx morphology. In order to describe the variability and significance of morpho-anatomical differentiation principal component analyses (PCA) and multivariate analyses of variances (MANOVA) was used. Canonical discriminant analysis (CDA) was used to test the hypothesis of morphological separation of the analyzed populations, and discriminant function analysis (DFA) was used to estimate the contribution of individual characters to the overall discrimination. Statistical analysis revealed three well differentiated group of populations.

**KEYWORDS:** *Teucrium montanum*, morphology, anatomy, leaf, differentiation

Poster presentation 27 02 50

### MORPHOLOGICAL CHARACTERIZATION OF *ANACAMPTIS* × *GENNARII*, A NATURAL ORCHID HYBRID NEW TO REPUBLIC OF MACEDONIA

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Hybridization is one of the leading mechanisms in plant evolution. Its wide presence among members of Orchidaceae family suggests it had a great role in the speciation of this plant group. *Anacamptis* × *gennarii* is a natural hybrid between *A. morio* and *A. papilionacea*. *A. morio* is perennial geophytes, distributed all over Euro-Mediterranean region, with great tolerance to different ecological conditions, which induce its wide morphological variability. Often inhabits poor grasslands and meadows, up to 2000 meters above sea level. *A. papilionacea* is widespread across Mediterranean, from the foothill of the Alps in the north, to the Caspian Sea in the east, appearing mainly on grasslands, garrigue and maquis on alkaline substrates, up to 1800 m above sea level. *A. × gennarii* was registered for the first time in Republic of Macedonia in 2016, on Pletvar Mountain Pass, in the vicinity of village Pletvar. Both parental taxa, as well as hybrid, were registered in thermophilous grassland (ass. *Astragalo-Helianthemum marmorei*) on dolomites limestone at 1008 m a.s.l. At the same locality, eight more orchid taxa were registered: *A. pyramidalis*, *A. laxiflora*, *A. coriophora* subsp. *coriophora*, *Cephalanthera damasonium*, *Neotinea tridentata*, *Ophrys scolopax* subsp. *cornuta*, *O. sphegodes* and *Platanthera chlorantha*. All three taxa (*A. morio*, *A. papilionacea* and *A. × gennarii*) were subjected to morphometric analyses. For these analyses, 30 parental (15 of each species) and 10 hybrid specimens were sampled. Only above-ground parts of the plants were sampled. A total of 36 morphometric characters were analyzed, as well as seven indexes derived from appropriate morphometric characters. Above-ground vegetative organs were measured with caliper, while dissected flowers were scanned and measured using Digimizer software ver. 5.3.2. Statistical analyses were done in Past ver. 3.20 and Statistica ver. 13.3. Based on these analyses, discriminant characters that distinguish hybrid taxa from parental are given. Specimens of *A. × gennarii* had in general intermediate values of measured characters with more or less similarities with one of the parental species, with the curvature and general appearance of spur being the main characters that distinguish this taxon from parental.

**KEYWORDS:** *Anacamptis morio*, *Anacamptis papilionacea*, morphometry, statistical analyses, Pletvar

Poster presentation 28 02 46

### FLOWER VARIABILITY AMONG *ORCHIS MASCULA* POPULATIONS FROM WESTERN AND CENTRAL AREA OF BALKAN PENINSULA

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The Early-purple orchid (*Orchis mascula*) is widely present throughout Europe in a variety of different habitats and altitudes. Its most apparent morphological characteristics are the three-lobed dotted labellum (lip) and its root system present in the form of two tubers. The focus of this paper were morphological analyses and comparison of flowers from 177 herbarized specimens from seven *O. mascula* populations, found in the area of the Balkan Peninsula, using linear morphometry. Plant material originated from Eastern and Western parts of the Republic of Serbia, Slovenia and Montenegro and kept in the Herbarium of the University of Novi Sad (BUNS). Flowers were removed from dried plant material, rehydrated, dissected and then scanned and measured in the software Digimizer ver. 4.5.2. A total of 25 morphometric characters were statistically evaluated. Statistical analyses were carried out using programs Past ver. 3.20 and Statistica ver. 13.3. Discriminant analysis (LDL) showed that the populations from Slovenia, Đerdap (NE Serbia), and Sopotnica (NW Serbia) are separated from the populations from Montenegro, while the population that originated from the Miroč Mt (NE Serbia) is of intermediate values in regard to the measured characters. Several previous research papers hypothesized that specimens (especially their flowers) of some taxa from genus *Orchis*, when living in sympatry with other food-deceptive orchid species, show more morphological differences when compared to the allopatric populations of the same species. In our case populations from Montenegro, which are sympatric with other food-deceptive orchids (*O. pauciflora*, *O. simia*, *O. quadripunctata*, *Anacamptis morio* subsp. *picta* and *Dactylorhiza viridis*) showed more morphological differences comparing to all other populations in allopatry. Further research, which will involve analysis of more populations from Balkan Peninsula, together with analyses of environmental conditions and habitat preferences of investigated species, will provide more information that supports our hypothesis.

**KEYWORDS:** Early-purple orchid, linear morphometry, food-deceptive orchid, sympatry, allopatry

Poster presentation 29 02 23

### THE SEED MORPHOLOGY OF GENUS *ATRIPLEX* SECTIONS *TEUTLIOIDES*, *STYLOSA* TAXA IN TURKEY

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The genus *Atriplex* L. includes 19 taxa belonging to five section section (Sect. *Teutlioides* Ulbrich, Sect. *Stylosa* Aellen, Sect. *Atriplex*, Sect. *Teutliopsis* Dumortier, and Sect. *Sclerocalymma* Ascherson) in Turkey. In this study, it was aimed to determine the seed morphology and seed coat micromorphology of 6 taxa belonging to two sections (*Teutlioides* and *Stylosa*,) in Turkey. Seed morphology is an important characteristic in the identification and classification of Chenopodiaceae family. The seed specimens were collected between 2011 and 2015 from different localities in Turkey. For LM, at least 50 seeds samples for each species were examined under the BAB-STR45 stereomicroscope with integrated camera system (BAB-DMC310, USB2.0 5v). For SEM, seeds were directly mounted on stubs and covered with gold. And then, seed surface was examined on SEM. In *Atriplex*, seeds are monomorphic, dimorphic or trimorphic. According to finding, both section *Teutlioides* and *Stylosa* have monomorphic seed structures. Section *Teutlioides* includes one taxon (*Atriplex halimus* var. *schweinfurthii*) and it has morphologically dark brown, orbicular, flattish (disk-like), radicle prominent, and surface rough seed structure. The seed coat surface ornamentation of taxon is reticulate-rugulate, radicle striate and longer than cotyledons. Section *Stylosa* includes two taxa (*A. lehmanniana*, *A. leucoclada*) and they have morphologically reddish, biconvex, comma shape lustrous seed structures. While *A. lehmanniana* has rugulate the seed coat surface ornamentation, *A. leucoclada* has psilate-rugulate ornamentation. But, in both species radicle is striate and longer than cotyledons. As a result, the micromorphology of seed coat of the sections was reported here the first time for Turkey and it has contributed to the taxonomy of the genus or sections.

**KEYWORDS:** Chenopodiaceae, *Atriplex*, seed morphology, sect. *Teutlioides*, sect. *Stylosa*

Poster presentation 30 02 10

**COMPARATIVE ANATOMICAL STUDIES ON STACHYS L. SECT. OLISIA DUMORT. SUBSECT. DISTANTES BHATTACHARJEE TAXA FROM TURKEY**

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This study was carried out in order to determine anatomical properties of *Stachys aleurites*, *S. bombycina* and *S. distans* var. *cilicica* belonging to *Stachys* sect. *Olisia* subsect. *Distantes*, which are spread in Turkey, and to reveal their taxonomic significance by comparing the properties of the taxa. The study material consisted of the samples collected from different parts of Turkey. Anatomical studies were carried out on stem, leaf and petiole cross sections and leaf surface sections of the specimens preserved in 70 % alcohol. The data obtained as a result of the studies showed that some anatomical characters, like the general appearance of the stem in cross-section, the state of the collenchyma tissue at the corners of the stem and the thickness of the leaf mesophyll, have diagnostic values in distinguishing species.

**KEYWORDS:** *Stachys*, anatomy, Turkey

Poster presentation 31 02 06

**COMPARATIVE MICROMORPHOLOGICAL STUDIES ON STACHYS L. SECT. OLISIA DUMORT. SUBSECT. DISTANTES BHATTACHARJEE TAXA SPREADING IN TURKEY**

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This study was carried out in order to determine palynological and nutlet micromorphological properties of *Stachys aleurites*, *S. bombycina* and *S. distans* var. *cilicica* belonging to *Stachys* sect. *Olisia* subsect. *Distantes*, which are spread in Turkey, and to reveal their taxonomic application by comparing the properties of the taxa. The study material consists of the samples collected from different parts of Turkey. Palynological studies were carried out using both light and scanning electron microscopy, with the material obtained from the collected herbarium samples. The data obtained as a result of the studies showed that some characters related to nutlet morphology had diagnostic value in distinguishing species

and that the characters related to pollen morphology were not useful in this respect. The nutlet morphological characters of taxonomic value are hairiness, winging state and general shape of nutlets.

**KEYWORDS:** *Stachys*, pollen, nutlet, Turkey

Poster presentation 32 02 36

**INFRAGENERIC GROUPING OF THE GENUS LIMONIUM MILL. (PLUMBAGINACEAE) IN TURKEY ASSESSED BY NUMERICAL TAXONOMY**

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The genus *Limonium* (sea lavenders) is included in the family Plumbaginaceae which has many uncertainties concerning primarily with its generic delimitation. Most of these uncertainties are linked to delimitation and circumscription of the large genera, such as *Acantholimon* and *Limonium*. The genus *Limonium* is the largest genus of the family with its center of diversity in the Mediterranean area. Infrageneric structure of the genus is quite complex and many small genera were segregated from it over the years. In the same time the numerous new species has been described in the recent past. This study examines the infrageneric grouping of the genus *Limonium* by using numerical taxonomic methods, based on 42 external vegetative and floral morphological characters, as well as habitat preferences of the 24 species recognized so far in Turkey. The results of this study suggest that there are basically six well defined sections in contrary to the flora and the sectional cut off line seems to lay at 0.82 similarity coefficient in the phenogram. Each of these sections clearly differ from the others based on their morphology and phytogeography.

**KEYWORDS:** Plumbaginaceae, *Limonium*, numerical taxonomy, Turkey

Poster presentation 33 02 44

**NEW TAXONOMICAL CONCEPT OF SILENE SAXIFRAGA GROUP (CARYOPHYLLACEAE) – THE BALKAN PENINSULA IS THE CENTRE OF TAXONOMIC DIVERSITY OF THE GROUP**

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Relationships among taxa belonging to or hypothesized to be closely related to *Silene saxifraga* group were mainly unresolved in previous classifications. Delimitation among taxa belonging to *S. saxifraga* group is discussed here on the basis of multivariate analyses of morphometric data and differences in qualitative characters as well as considering phylogenetic relationships outlined in recent molecular studies. Samples from 122 populations belonging to 22 taxa, including *S. linoides*, *S. multicaulis* and *S. waldsteinii*, were analysed. Ten taxa are treated as species (*S. ancræe*, *S. campanula*, *S. conglomeratica*, *S. linoides*, *S. multicaulis*, *S. orphanidis*, *S. paeoniensis*, *S. pindicola*, *S. saxifraga*, *S. schmuckeri*, *S. triflora*, *S. velcevii* and *S. waldsteinii*), with eight subspecies within *S. saxifraga* (*S. s.* subsp. *antri-jovis*, *S. s.* subsp. *dirphyia*, *S. s.* subsp. *lurensis*, *S. s.* subsp. *notarisii*, *S. s.* subsp. *pirinica*, *S. s.* subsp. *saxifraga*, *S. s.* subsp. *taygetea* and *S. s.* subsp. *vourinensis*). One new species (*S. ancræe*) and one new subspecies (*S. saxifraga* subsp. *lurensis*) are described, whereas six taxa are given as new combination and five out of them as new status. The most reliable characters for species delimitation are qualitative and meristic; in particular habitus features, indumentum and the number of flowers. Subspecies delimitation requires combination of several morphological characters but also ecological and chorological data. Eight out of ten taxa treated as species within revised *S. saxifraga* group as well as six out of eight subspecies within *S. saxifraga* are endemic for the Balkan Peninsula. The highest number of taxa is recorded in gorges and canyons below 500 m a.s.l. and the number of species declines along the vertical gradient. The most widespread taxa, *S. pindicola* and *S. saxifraga*, also have the widest elevation range. Subspecies within *S. saxifraga* are mostly local endemics of single mountains. Four species are recorded only on carbonate substrate, whereas two subspecies are obligate ultramafic endemics. Investigated taxa are mostly recorded on cliffs and rocky grounds.

**KEYWORDS:** Balkan Peninsula, chorology, diversification, ecology, *S. saxifraga* group, taxonomy

Poster presentation 34 02 37

**TAXONOMY OF YELLOW FLOWERED SEMPERVIVUM SPECIES IN BALKAN PENINSULA: INSIGHT BASED ON EVIDENCE ON LEAF EPIDERMAL STRUCTURES**

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In the Central, Eastern and the Southern parts of Balkan Peninsula a two group of yellow flowered *Sempervivum* species can be distinguished. The first group, provisionally named here as „*Sempervivum ciliosum* complex“, includes taxa: *S. ciliosum* Craib, *S. galicicum* (A.C.Sm.) Micevski, *S. klepe* Micevski, *S. jakucsii* Pénzes and *S. octopodes* Turill, while second group („*Sempervivum ruthenicum* complex“) includes taxa: *S. leucanthum* Pančić, *S. kindingeri* Adamović and *S. zeleborii* Schott. The intrageneric taxonomy of these groups is blurred as a consequence of a small number of studies encountered within these taxa as well as highly expressed phenotypic plasticity of this species. As a result, it is not possible to certainly establish whether all the taxa are well-defined species. Main objectives of this study are the analysis of the trichomes on surface and margin of rosette leaves, which can be potentially used as taxonomic characters. Plant material was collected from the field and cultivated under the same conditions for a period of minimum one year. Trichomes were analysed using light microscope and stereo microscope within fifteen populations (a total of 225 individuals). Measuring of quantitative characters of trichomes was carried out in software Digimizer 8.0. The results show the presence of glandular trichomes with multicellular, biseriate stalk and multicellular (rarely unicellular), biseriate head in all examined populations, both on abaxial and adaxial surface of rosette leaves. Excluding sporadic presence of stellate trichomes, the presence of non-glandular hair has not been established. On margins of the leaves are also established only glandular trichomes with large extent of differences in length among examined taxa. Hairy indumentum was represented in all examined individuals. Although there are no major differences in the type of trichomes the largest differences were observed in their length and density within examined taxa, which can be used as tentative taxonomic characters.

**KEYWORDS:** *Sempervivum*, trichomes, indumentum, rosette leaves, variability, Balkans

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### KARYOLOGICAL VARIATION IN *PROSPERO AUTUMNALE* (L.) SPETA (HYACINTHACEAE) COMPLEX IN PANNONIAN PLAIN AND BALKAN PENINSULA

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*Prospero autumnale* (L.) Speta (Hyacinthaceae) is a karyologically highly variable complex with three base chromosome numbers ( $x = 5, 6, 7$ ), four diploid cytotypes (AA, B<sup>5</sup>B<sup>5</sup>, B<sup>6</sup>B<sup>6</sup> and B<sup>7</sup>B<sup>7</sup>), each with unique combination of genome size and chromosome number. Cytotype B<sup>7</sup>B<sup>7</sup> is widespread in whole Mediterranean, while the remaining three cytotypes are more geographically restricted to Iberian Peninsula and Morocco (AA), Libya (B<sup>5</sup>B<sup>5</sup>) and Crete (B<sup>6</sup>B<sup>6</sup>). Three of the diploid cytotypes of the complex hybridize and polyploidize giving rise to a wide range of auto- and allopolyploids. In addition, this circum-Mediterranean complex is characterized by frequent presence of B chromosomes and supernumerary chromosomal segments (SCSs), as well as variation in genome size both between and within the cytotypes. The present study focuses on the analyses of the variation of genome size and ploidy levels in 17 populations of *P. autumnale* complex in the Pannonian Plain and Balkan Peninsula. Chromosome numbers have been established in root tips meristems following Feulgen staining and fresh leaves were used for measurements of genome size. Genome sizes were measured using propidium-iodide (PI) flow cytometry. Chromosome counts were obtained for 36 individuals and four ploidy levels were detected in the study area. Diploids ( $2n=2x=14$ ) and tetraploids ( $2n = 4x = 28$ ) were found in five populations each, while hexaploids ( $2n = 6x = 42$ ) in seven populations. The genome sizes were measured for 56 individuals and the 1C-values ranged between 4.17 and 4.59 pg in diploids, 7.64 and 9.41 pg in tetraploids and between 10.41 and 11.76 pg in hexaploids. The mean values of monoploid genome size (1Cx) of diploids (4.37 pg), tetraploids (4.07 pg) and hexaploids (3.68 pg) showed that 1Cx-values decrease with increasing ploidy levels. Both diploids and tetraploids exhibited differences in 1Cx-values between populations, while such difference between hexaploid populations was not statistically significant.

**KEYWORDS:** chromosome number, flow cytometry, ploidy levels, C-value

Poster presentation 36 02 49

### GAGEA PRATENSIS S. LAT. (LILIACEAE) IN THE PANNONIAN BASIN: CYTOGEOGRAPHY AND CYTOTYPE COEXISTENCE ON DIFFERENT SCALES

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Due to an extensive polyploidy and morphological variability, *Gagea pratensis* s. lat. has been considered to be taxonomically complicated group. Together with *Gagea pusilla* (regarded here as closely related to *G. pratensis* s. lat.), three ploidy levels have been reported from Central Europe, i.e. diploids ( $2n=24$ , *G. pusilla*), tetra- and pentaploids ( $2n=48$  and  $2n=60$ , *G. pratensis* s. lat.). We aimed to explore cytotype diversity and patterns of cytotype distribution of *G. pratensis* s. lat. (incl. *G. pusilla*) in eastern part of Central Europe with special emphasis on the Pannonian Basin from where all three ploidy levels were reported previously. We also focused on assessing patterns of cytotype coexistence on both large (i.e. landscape) and small (i.e. locality) scales. Altogether, we studied 1875 plants from 165 populations in six countries. Ploidy level of plants was established using flow-cytometry and standard protocol of sample preparation with DAPI staining. We found five cytotypes occurring in the studied region: apart from previously known diploids, tetraploids and pentaploids, we also detected triploids and hexaploids. Diploids were confined mainly to the lowlands. Triploids were of two types: probable autotriploids that are morphologically close to *G. pusilla* and were rarely recorded in otherwise diploid populations and probable allotriploids that are morphologically similar to tetraploid *G. pratensis* s. lat. The allotriploids were found in mixed populations of diploids and tetraploids. Tetraploids were confined mainly to xeric sites and represent the most common cytotype in lowlands east of the Danube river, whereas penta- and hexaploids were found also in higher altitudes as well as in rather mesophilous regions. We found 30 mixed populations altogether, mainly with diploid-tetraploid, tetraploid-pentaploid and pentaploid-hexaploid co-occurrence. Preliminary sampling on small scale showed that triploids in otherwise diploid populations (i.e. probable autotriploids) are very rare (0.6% out of 628 analysed plants from two populations), whereas triploids in diploid-tetraploid mixed populations (i.e. probable allotriploids) are more common (11% out of 156 analysed plants in one population). Our ploidy screening revealed distinct distribution patterns in *Gagea pratensis* s. lat. and it will serve as a basis for further research on the evolutionary processes and taxonomic assessment within this complicated group.

**KEYWORDS:** flow-cytometry, *Gagea pusilla*, mixed-ploidy populations, polyploidy

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### DIPLOID *HIERACIUM* AND *PILOSELLA* SPECIES (ASTERACEAE) ON THE BALKAN PENINSULA

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*Hieracium* and *Pilosella* (Asteraceae) are notorious for the taxonomic difficulties which are due to the co-occurrence of normal sexual reproduction with hybridization, polyploidy and apomixis. In fact, both genera form two separate large agamic complexes. In such complexes diploid sexual species are crucial for understanding the morphological variation and taxonomy of the whole groups. The objective of this study is to review the currently known diploid species in *Hieracium* and *Pilosella* on the Balkan Peninsula, to outline their distribution and to discuss the consequences for the taxonomy of the respective taxonomic groups. The results are based on personal studies of the Balkan *Hieracium* and *Pilosella* species by both authors as well as on a literature survey of published chromosome counts. The ploidy level of the personally studied taxa has been determined by direct chromosome counting in squashes of haematoxylin-stained root-meristem or by flow cytometry using fresh leaves. The results suggest that the Balkan Peninsula harbours more than 20 *Hieracium* and *Pilosella* species in which diploid counts have been recorded, for example, *Hieracium cernuum*, *H. heterogynum*, *H. kittaniae*, *H. naegelianum*, *H. petrovae*, *H. plumulosum*, *H. renatae*, *H. sabaudum*, *H. sparsum*, *H. transylvanicum*, *H. umbellatum*, *Pilosella hoppeana*, *P. onegensis*, *P. pavichii*, *P. petraea*, *P. rhodopaea*, *P. pseudopilosella*, *P. serbica*. The poster presents data about the geographic distribution of the karyologically studied species and populations, and comments on their taxonomic affiliation and treatment. Apparently, the Balkan Peninsula is the richest in diploid *Hieracium* and *Pilosella* species area in Europe.

**ACKNOWLEDGEMENTS:** The studies have been partly supported by the Bulgarian National Science Fund under the project 'Flora of the Republic of Bulgaria, vol. 12: Biological Diversity in Asteraceae subfam. Carduoideae and Cichorioideae', project contract DN01/7.

**KEYWORDS:** agamic complexes, Asteraceae, Balkan Peninsula, *Hieracium*, *Pilosella*, diploid species

Poster presentation 38 02 45

### TWO DIFFERENT EXAMPLES OF HYBRIDIZATION IN THE GENUS *CYANUS* FROM GREECE

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*Cyanus* sect. *Protocyanus* includes cca 67 perennial species distributed in north-western Africa, Europe (except northern parts) and western Asia. Nowadays, 18 species of this section are reported from the Balkan Peninsula. During field research in 2010-2012 we found morphologically intermediate plants at two localities in Greece where genetically well-defined species, namely *Cyanus epirotus* with *C. pichleri* and *C. pindicola* with *C. tuberosus* s.l., occurred in sympatry. Their hybridization has not been documented previously. In this study, we included individuals collected across the ranges of parental species supplemented with detail sampling from supposed hybrid localities. Potentially hybrid origin of intermediate individuals was studied using AFLP markers, flow cytometry and multivariate morphometrics. Our data confirm hybridization at both localities, however there are substantial differences. In the first case, parental species differ in chromosome number, *C. pichleri* is tetraploid ( $2n=4x=40$ ) and *C. epirotus* is diploid ( $2n=2x=20$ ). Their hybrid as result of heteroploid hybridization has expected triploid ploidy level ( $2n=3x=30$ ) and is morphologically intermediate AFLPs show only minute genetic variation of its individuals, therefore prevalence of clonal spreading is expected. In the second case, parental species do not differed in ploidy, *C. pindicolus* as well as *C. tuberosus* s.l. are diploid ( $2n=2x=20$ ). Their homoploid hybridization has resulted in formation of a hybrid swarm that consist of plants performing various level of genetic and morphological intermediacy between the two parental species. It seems that there is only weak breeding barrier between parental species and their hybrids what enables their recurrent backcrossing.

**ACKNOWLEDGEMENTS:** Study was financially supported by the Scientific Grant Agency of the Slovak Republic (VEGA 2/0096/15).

**KEYWORDS:** AFLPs, *Centaurea*, flow cytometry, homoploid hybridization, heteroploid hybridization, multivariate morphometrics

### DIFFERENCES IN CLONAL GROWTH MAY EXPLAIN THE DOMINANCE OF POLYPLIIDS IN DIPLOID-POLYPLIID PRIMARY CONTACT ZONES

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Polyploidy has long been recognized as a significant force in the evolution of plants. It is known that genome duplication can induce changes in gene expression, ecophysiology, morphology and/or life-history traits. Among other traits, clonality is associated with the incidence of polyploidy in vascular plants, however only few studies tested the assumption that transition to polyploidy might increase clonal reproduction. In the present study, we aim to test whether clonal growth is greater in polyploid plants than in diploids using the *Pilosella rhodopea* (Asteraceae) as a model system. This species encompasses several cytotypes (from 2x to 6x) which occur in numerous mixed-ploidy populations. Triploids are the most frequent cytotype, followed by tetraploids and diploids, whereas 5x and 6x are rare. Polyploids are of autopolyploid origin and arise repeatedly in primary contact zones. All cytotypes reproduce sexually, mate freely and are at least partially fertile, although seed set of polyploids is severely reduced. Moreover, a significant shift in proportion of cytotypes was found in seed progeny, where polyploids and especially triploids are extremely rare, what is in strike contrast to their predominance in the field. Besides sexual reproduction, all cytotypes are able to spread also *via* underground stolons. Increased clonal growth of polyploids in comparison to their diploid counterparts could facilitate the establishment of rarely formed polyploids and thus explain their distributional success. The extent of clonality in diploids and polyploids of *P. rhodopea* was studied through (i) an analysis of spatial distribution and size of clones in two natural populations using multilocus genotyping (SSRs) and (ii) a quantification of accessory rosettes production and incidence of flowering in the greenhouse experiment. Our results showed that autopolyploidization induced vegetative growth, which enables the establishment and effective clonal spreading of rarely formed and less fertile polyploids and thus underlies their supremacy in natural populations.

**KEYWORDS:** clonality, greenhouse experiment, microsatellite genotyping, *Pilosella*, polyploidy

### LONG NEGLECTED DIVERSITY IN THE ACCURSED MOUNTAINS: RANUNCULUS BERTISCEUS IS A GENETICALLY AND MORPHOLOGICALLY DIVERGENT SPECIES

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Southern European mountain ranges such as the Pyrenees, Alps, Apennines, Carpathians and Dinaric Alps have long been recognized as important hotspots of genetic diversity and areas of high endemism. Reflecting the geographical complexity of these mountain ranges as well as the complex geotectonic events and climatic fluctuations, many European high-mountain species exhibit disjunctions on a variety of geographical scales. One of the long neglected, poorly investigated and unresolved taxonomic problem concerns Apennine and Balkan members of *Ranunculus* sect. *Leucoranunculus*. This section comprises small, mostly one-stemmed plants with one to two (rarely more) flowers, petiolate basal leaves with a more or less lobed suborbicular or orbicular blade and obscure venation, and white honey leaves. According to the most recent taxonomic concept, this section includes *R. crenatus* distributed predominantly in siliceous massifs of the Carpathians and the Balkan Peninsula, with a highly disjunct partial distribution area in the Eastern Alps, and *R. magellensis*, which is considered a calcicolous endemic to the Central Apennines. Alternatively, *R. magellensis* was suggested to occur also in carbonate ranges of the Albanian Alps (Accursed Mountains / Alpet Shqiptare / Bertiscus / Bjeshkët e Nemuna / Prokletije), which would render this species amphii-Adriatic. We used complementary molecular methods – sequences of the nuclear ribosomal and the plastid region along with amplified fragment length polymorphisms, relative genome size measurements and morphometric analyses to elucidate the relationships within *Ranunculus* sect. *Leucoranunculus*, specifically between *R. magellensis* from the central Apennines and populations of more widespread *R. crenatus* from the Alps, Carpathians and the Balkan Peninsula. The results of our study showed that populations growing on limestone in the Albanian Alps in Northern Albania and southern Montenegro are divergent and should be recognised as a new species, *Ranunculus bertisceus*. These populations differ not only from *Ranunculus crenatus* s. s., which grows on silicates, but also from the Apennine endemic *R. magellensis*. It is interesting to note that the Styrian populations of *Ranunculus crenatus*, which occur very locally in the Niedere Tauern area,

### A NEW ENDEMIC SPECIES FROM NORTH AFRICA, GONIOLIMON AFRICANUM (PLUMBAGINACEAE)

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*Goniolimon africanum* is described and illustrated as a new endemic species from North Africa (Algeria and Tunisia). Ernest Cosson collected this species for the first time during botanical excursion in Algeria in May 1852 and gave a provisional name '*Goniolimon luteolus*' nom. nud. According to previously published data and herbarium specimens from the European herbaria, only *G. tataricum* was recorded in the African flora. A comparative morphological analysis was carried out among individuals of African populations and several related European *Goniolimon* species and it was concluded that African populations are morphologically well distinguish from the European ones. Based on literature and herbarium data, we present distribution, ecology characteristics as well comparative diagnostic features of related European taxa (*G. tataricum*, *G. incanum*, *G. italicum*, *G. dalmaticum*, *G. heldreichii*, *G. sartorii*, and *G. bessenianum*) and new endemic species *G. africanum* from North Africa.

**KEYWORDS:** Algeria, *Goniolimon africanum*, herbaria, morphology, taxonomy, Tunisia

originate from immigration from the Bosnian mountains and not from the Western Carpathians as previously assumed.

**KEYWORDS:** Accursed mountains, Amphii-adriatic disjunction, Balkan Peninsula, endemism, *Ranunculus crenatus* s.l.

### HYBRIDIZATION IN SESLERIA (POACEAE) – NEW HYBRID REVEALED BY MOLECULAR AND MORPHOMETRIC ANALYSES FROM KOMOVI MTS. IN MONTENEGRO

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*Sesleria* (Poaceae) is a species-rich and taxonomically difficult genus, distributed throughout western Eurasia and North Africa, with highest diversity on the Balkan Peninsula. A recent phylogenetic study of *Sesleria* and closely related genera has shown that, from an evolutionary point of view, hybridization coupled with polyploidisation was likely responsible for diversification within the genus. During our fieldtrips to the Komovi mountain range in Montenegro, individuals morphologically intermediate between *S. coeruleans* and *S. interrupta* were observed. In order to investigate the occurrence of hybridization between the two species, we used amplified fragments length polymorphism (AFLPs) as well as morphometric analyses of reproductive and vegetative organs. The results of molecular analyses confirmed the existence of a hybrid individuals that has intermediate morpho-anatomical characteristics and is ecologically divergent from the parental taxa.

**KEYWORDS:** AFLP, hybridization, morpho-anatomy, *Sesleria*, Poaceae

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### GONIOLIMON TATARICUM (PLUMBAGINACEAE) AND ITS RELATIVES FROM SE EUROPE AND THE APENNINE PENINSULA BASED ON MORPHOLOGICAL AND MOLECULAR EVIDENCE

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The vast majority of the 20-30 species belonging to the genus *Goniolimon* are components of the steppe or steppe like rocky vegetation. Eleven *Goniolimon* species are listed in the European flora and the main parts of distribution ranges of majority of them are in the Ukraine and the European part of Russia. Six *Goniolimon* species are currently found in the Balkans, and they comprise *G. besserianum*, *G. dalmaticum*, *G. heldreichii*, *G. incanum*, *G. sartorii* and *G. tataricum*. In order to provide new insights into the origin and evolutionary history of *Goniolimon* species in the Balkans, we analysed variability of two maternally inherited plastid intergenic spacers (*rpl32-trnL* and *3'rps16x2F2-5'trnK*) in 110 individuals belonging to six *Goniolimon* species from the Balkans and to *G. italicum* from the Apennines. In order to resolve ambiguities related to the relations and current taxonomic treatments of three closely related taxa, *G. tataricum*, *G. dalmaticum* and *G. italicum*, we assessed variability of 14 morphological characters in 707 individuals belonging to these taxa. We provide a new taxonomic treatment for *Goniolimon* with description of three new *G. tataricum* subspecies and demonstrate that the history of *G. tataricum* at the westward range-edge of its current distribution was rather complex and driven by geo-historic events in this region. These events facilitated multiple waves of expansions of lineages originating from sources outside of the Balkan Peninsula and also of those emerging subsequently in this region over time and periodically bursting throughout more localized areas during the Pleistocene. An initial spread of an ancient *G. tataricum* lineage throughout SE Europe probably occurred during the Messinian Salinity Crisis. Frequent inter- and intraspecific hybridization/introgression as well as retention of ancestral polymorphisms hampered our phylogenetic analyses and thus further studies are required to fully resolve relations and evolutionary history of *G. tataricum* and related taxa.

**KEYWORDS:** Apennine and Balkan Peninsulas, divergence time estimates, *Goniolimon*, morphometry, phylogeny, taxonomy

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### ON THE DIANTHUS FRUTICOSUS AND DIANTHUS JUNIPERINUS GROUPS IN GREECE: DISTRIBUTION PATTERNS, FUNCTIONAL DIVERSITY AND PHYLOGENETIC RELATIONSHIPS

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Caryophyllaceae is a large family consisting of approximately 2200 species (herbs and subshrubs) traditionally placed in some 70 to 86 genera. *Dianthus* is the second largest genus within the Caryophyllaceae family, with an estimated number of 300 species most of which occur in SE Europe, SW Asia and Africa. The *Dianthus* clade contains a high number of endemic species with often very restricted distribution ranges, which probably are the result of a rapid evolutionary radiation. Based on recent studies, the genus *Dianthus* in Greece includes 87 taxa distributed in 44 species, of which 39 taxa (48.6%) are Greek endemics and 53 taxa are characterized as Range-restricted. The current research focuses on two endemic species groups, the *Dianthus fruticosus* group (with 8 subspecies considered to be parapatric) and the *Dianthus juniperinus* group (with 7 subspecies considered to be allopatric). *Dianthus fruticosus* is widespread in the S Aegean area, Ionian Islands, south Peloponnisos and occurs on coastal vertical, limestone rocks. *D. juniperinus* is restricted to the island of Crete and occurs in crevices and ledges of limestone rocks. Locality data (geographical coordinates) have been used for the analysis of their spatial patterns within the phytogeographical regions of Greece, along a geographical and altitudinal gradient. Specimens of these two groups but also other members of *Dianthus* are currently collected in the field to serve for molecular analysis, as well as cytology and morphology. In a first step, sequences of multiple plastid and nuclear regions will be included into a large scale phylogenetic overview to test the monophyly of the two species groups. High quality DNAs will further be used to develop a phylogenomic marker system to illuminate species level-relationships and species limits. A synopsis of all the reviewed literature on all the aspects of this research is presented to summarize the current knowledge and a working-level taxonomic backbone of these taxa, but also to specify hypotheses and questions. With the aim to assess the conservation status and the risk of extinction of the studied taxa by examining different climatic scenarios in combination with species distribution data, recent methodological approaches have been reviewed and will be presented.

**KEYWORDS:** *Dianthus*, *juniperinus*, *fruticosus*, phylogeny, species delimitation

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### PHYLOGENETIC AND MORPHOLOGICAL STUDIES OF A LESS KNOWN GENUS FRORIEPIA (APIACEAE)

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Apiaceae is one of the largest families by ca. 455 genera and ca. 3700 species, especially distribute in temperate region. Although the taxa of the family are the best known because of their characteristic inflorescences, fruits and chemical contents, hitherto micromorphological characters and phylogenetic placement of *Froriepia* C. Koch are unknown. The genus is represented by two species: *F. gracillima* Leute is endemic for Turkey and only known from type locality which is very close to Iran border in eastern of Turkey. *F. subpinnata* (Ledeb.) Baill. is distributed in Caucasia and north western of Iran. The difference of these two species is mainly that *F. gracillima* has very short habit. While it is less than 10 cm, *F. subpinnata* is more than 50 cm. In the current research, we comparatively studied morphological characters of these two species. We examined micromorphology of the pollen exine and mericarp surface by scanning electron microscope for the first time; also presented distribution map, detailed micrographs and photographs. The pollen grains of both species have rugulate exine ornamentation. While *F. subpinnata* has distinctly ridged mericarps, *F. gracillima* has weak ridged ones. Additionally, we analyzed using bayesian and maximum likelihood approaches based on internal transcribed spacer (ITS) and matK sequences to determine its phylogenetic placement in the family.

**KEYWORDS:** *Froriepia*, Apiaceae, endemic, morphology, Turkey, Iran

Poster presentation 46 02 41

### EDRAIANTHUS TARAE (CAMPANULACEAE), AN INTRIGUING TAXON OF THE BALKAN PENINSULA - EVIDENCE FROM MORPHOMETRIC, MOLECULAR AND GENOME SIZE STUDIES

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The taxonomically intricate *Edraianthus tarae*, a strictly endemic plant from the canyon of river Tara is reviewed using morphological, molecular and genome size data. Several morphological features separate this species from typical members of *Edraianthus*. First, the flowers are white and unusually large (3–5 cm long). More importantly, flowers are pedicelate and are arranged in terminal dichasia or polychasia. This inflorescence type is unusual not only in comparison with other *Edraianthus* species but also represents a curiosity for the Campanulaceae as a whole. Finally, the fruit dehiscence represents perhaps the most distinct feature found in *E. tarae*. Its capsules open by 3–4 irregular pores at the calyx base, as opposed to the lateral openings found in campanuloid taxa or apical found in all other members of the genus *Edraianthus*. For all this reasons, this taxon was segregated into its own genus, *Protoedraianthus*. Our formal morphometric analysis clearly indicate its morphological distinctiveness in relation to all other representatives of the genus *Edraianthus*, supporting the idea of segregation of this species into its own genus. In contrast to morphometric analysis, our phylogenetic analyses based on plastid DNA (*trnL-F* region and *rbcL-atpB* spacer) and nuclear nrETS sequences confirm the monophyly of *E. tarae* and revealed that it makes a natural group with *Edraianthus* s.str., and that it belongs to *E. graminifolius*-complex, the largest and most diverse group of *Edraianthus*. Within this group, *E. tarae* (= *Protoedraianthus tarae*) forms an isolated lineage, positioned as a sister group to the rest of the *E. graminifolius*-complex. The genome size analysis largely correspond with molecular results, indicating that, despite the morphological distinctiveness, *E. tarae* (= *Protoedraianthus tarae*) is clearly nested within *Edraianthus* and should be treated as congeneric with it. Finally, results of our morphometric, molecular and genome size analysis allow us to confirm *Edraianthus tarae* as stenoendemic species of canyon of river Tara in Montenegro. Given that names *Edraianthus tarae* and *Protoedraianthus tarae* were not validly pub-

lished, respective nomenclatural and taxonomical standings are discussed and reevaluated. The name *Edraianthus tarae* is validated, a new differential diagnosis, description and illustration are provided, and its conservation status is assessed.

**KEYWORDS:** Balkan Peninsula, *Edraianthus*, endemic taxa, genome size, morphometry, nomenclature, plastid and nuclear sequences

Poster presentation 47 02 35

### GENETIC AND MORPHOLOGICAL DIFFERENTIATION WITHIN THE BROADLY DEFINED *CAMPANULA VERSICOLOR* (CAMPANULACEAE)

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*Campanula versicolor*, commonly known as various-colored bell-flower, is a constituent of the *C. pyramidalis* complex, distributed in the southern Balkan Peninsula with a small disjunct range in the southeastern Italy. Morphologically, it is highly variable species, which lead to the description of sixteen taxa which can be related to it. However, in modern floristic literature and checklists those taxa are neglected and considered as synonyms of broadly defined *C. versicolor*. Our extensive population sampling covered the entire range of the taxonomically intricate *C. versicolor* s.l. Additionally, populations of recently described taxa from Albania – *Campanula korabensis*, *C. korabensis* subsp. *bicajensis* and *C. longipetiolata* are included in this study as well, based on the description from protologues, type materials and personal observations. Reconstruction of relationships among the populations based on microsatellite DNA data revealed three genetic groups of populations, which are morphologically and geographically well differentiated. Based on the obtained results a new taxonomic treatment of *C. versicolor* s.l. was proposed, along with the key for the identification, description, distribution and habitat data.

**KEYWORDS:** *Campanula pyramidalis* complex, *C. versicolor*, microsatellites, morphometrics, taxonomy

Poster presentation 48 02 34

### DISENTANGLING RELATIONSHIPS BETWEEN AMPHI-ADRIATIC *EUPHORBIA SPINOSA* AND BALKAN ENDEMIC *E. GLABRIFLORA* (EUPHORBIACEAE)

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*Euphorbia spinosa* and *E. glabriflora* are shrublets distributed in the Mediterranean region. The former is widespread along the coasts of the Ligurian, Tyrrhenian, Ionian and Adriatic Sea, whereas the latter grows in continental regions of the western and southern Balkan Peninsula, mostly on serpentines. Nuclear ribosomal internal transcribed spacer (ITS) sequences indicate that both species together form a monophyletic group closely related to the *E. acanthothamnus* alliance, but the relationships within the clade remain unresolved. Amplified fragment length polymorphism (AFLP) fingerprints indicate a northwest-southeast trend in genetic differentiation, with two weak groups indicated by Neighbor-Joining and NeighbourNet analyses. The two groups include populations from France, Italy, Croatia and Bosnia and Herzegovina on one hand and populations from Montenegro, Serbia, Albania, Macedonia and Greece on the other hand. Bayesian Analysis of Populations Structure (BAPS) and K-means clustering of AFLPs revealed three clear allopatric groups of populations. Two groups correspond to groups revealed by the Neighbor-Joining analysis, and the third group is distributed in the transition area between the two before-mentioned groups in Serbia, Montenegro, northern Albania and northern Macedonia. Morphometric analyses showed no significant differentiation between the genetic groups and only slight differentiation between both taxa. This is likely a result of many morphologically transitional forms, which can be found especially in the area of the intermediate genetic group. The taxonomic status of *E. glabriflora* thus needs to be reconsidered.

**KEYWORDS:** AFLP, *Euphorbia spinosa*, *E. glabriflora*, morphometrics, ITS

Poster presentation 49 02 21

### MOLECULAR PHYLOGENY OF THE GENUS *AURINIA* (BRASSICACEAE) – PRELIMINARY REPORT

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The Balkan peninsula is one of the major European biodiversity hotspots which is considered as a result of a series of glaciations through Pleistocene. The alternating periods of glacials and interglacials lead to climate changes which re-shaped entire environments and their associated biological communities. Due to its geographical position the impact of the glaciations was not so severe on the Balkan Peninsula making it a perfect refugium for many species which are often endemic or disjunctly distributed. In order to better understand how the climate changes lead to the genetic biodiversity we conducted a phylogenetic study of the endemic south – eastern European plant genus *Aurinia*, a genus within the Alysseae tribe (Brassicaceae). Morphologically, the species within the genus are perennial herbs with an indumentum of stellate hairs, sinuate or dentate rosetteleaves with petioles grooved and with swollen bases persistent on caudices and raceme inflorescences which are exclusively yellow-flowered. The objective of the study was to determine the phylogeny of the genus and to delimit the spatial boundaries of all taxa in the genus. Consequently, the obtained data will provide insights into spatiotemporal diversification of the genus and offer an outline for the reconstruction of individual *Aurinia* phylogeographies. Sequence data from the nrDNA ITS (nuclear ribosomal DNA internal transcribed spacer) and a plastid region of the *ndhF* gene (NADH dehydrogenase F) were obtained and used in phylogenetic analyses. The nuclear dataset mostly showed congruence with the current taxonomy, as well as the formation of geographically defined groups within some taxa. Analyses of plastid dataset yielded a network showing geographical structuring and lacking clear taxonomical pattern. The observed incongruences between nuclear and plastid datasets are probably a result of hybridization or incomplete lineage sorting and reflect the complex evolutionary history of *Aurinia* taxa within microrefugial areas on the Balkan peninsula.

**KEYWORDS:** *Aurinia*, Balkan peninsula, ITS, *ndhF*, phylogeny

Poster presentation 50 02 20

### A PRELIMINARY MOLECULAR SYSTEMATIC STUDY ON TURKISH *PRUNUS* L., SUBGENUS *CERASUS* (Mill.) Focke

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*Prunus* subgenus *Cerasus* comprises more than 100 taxa of which some are economically important like *P. cerasus*, *P. avium* and *P. mahaleb*. Subgenus *Cerasus* is divided into eight sections based on their morphological traits. Although Turkey is one of the centers of diversity of subgenus *Cerasus*, there is no molecular study on the individuals from this area. Our study therefore is a preliminary work aimed to infer (i) the phylogenetic relationships of the Turkish subgenus *Cerasus* species, (ii) the genetic diversity, and (iii) to clarify the unresolved taxonomic rank of some of these taxa. We analyzed 55 individuals from different populations throughout Turkey representing 12 taxa from three sections using two chloroplast markers (*matK*, *ycf1*). Seven different haplotypes were identified within our data set. A parsimony network approach showed up to three different chloroplast haplotypes existing within a single species and up to four species sharing single haplotypes. Incomplete lineage sorting occurred in four taxa of Section *Microcerasus*. The lowest diversity was observed in cultivated species and probably results from selection by humans.

**KEYWORDS:** *Prunus*, *Cerasus*, phylogeny, *matK*, *ycf1*, Rosaceae

Poster presentation 51 02 19

### PHYLOGENETIC RELATIONSHIPS OF ANNUAL SPECIES OF THE GENUS *ALYSSUM*

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Genus *Alyssum* (Brassicaceae), since the last major taxonomic revision, comprises 114 species which were traditionally placed into two sections (*Alyssum* sect. *Alyssum* and *A. sect. Psilonema*). Twenty eight of them are annual herbs with native distribution in Eurasia and North Africa. Previously published molecular studies based on ITS and chloroplast DNA markers (using direct sequencing) did not show clear phylo-

genetic resolution of taxa and suggested that the two sections are not supported. Five ploidy levels were previously reported for the annual species. In the present study, we are trying to reveal the phylogenetic relationships and evolutionary history of annual taxa of *Alyssum* using one chloroplast and two nuclear molecular markers (including the cloning of PCR products to detect within-individual allelic polymorphism). Our field samples include 16 species and 230 population samples from the Apennine, Balkan and Iberian Peninsulas, Central Europe and Morocco. Ploidy levels of plants were determined by chromosome counting and flow cytometry. We employed nuclear ITS (Internal Transcribed Spacer) and chloroplast DNA (*rpoB-trnC*) markers firstly on data set of 81 populations (162 specimens) and nuclear DET1 (De-Etiolated homolog 1) marker on selected group of 25 populations (50 specimens). Preliminary results reveal the relationships of several little explored taxa, including species with assumed hybrid origin.

**KEYWORDS:** *Alyssum*, annual species, phylogeny, molecular makers, polyploidy

Poster presentation 52 02 63

### DO GENETIC DATA SUPPORT THE PRESENCE OF FAGUS MOESIACA IN THE CENTRAL BALKANS (SERBIA)?

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European or common beech, *Fagus sylvatica* (L.), is one of the widely distributed and thoroughly investigated European tree species present also in the Balkan Peninsula. However, forest authorities in Serbia support the presence of *Fagus moesiaca* [Domin, Malý (Czech.)] in this central Balkans country. Along with the fact that this represents an important botanical issue, it also has severe consequences to the Serbian economy, i.e., to the reproductive material trade because Serbia cannot export the seeds of *F. moesiaca* to countries that import *F. sylvatica* seeds. Furthermore, the existence of *F. moesiaca* in the Balkans has been questioned by several authorities, and, in addition, several large-scale molecular genetics studies over the past decade failed to detect genetic difference among *F. sylvatica* individuals from Europe and those from the central Balkans delineated as *F. moesiaca*. These studies, however, included a rather limited sample from the central Balkans. In order to address *F. sylvatica*/*F. moesiaca* problem relevant for Serbia from both scientific and economic aspects, we carried out genotyping of 192 beech individuals from 13

populations from Serbia with 12 nuclear and four chloroplast microsatellites. We present here first outcomes of our study and demonstrate that beech populations in Serbia and indeed genetically structured. The analyses of beech individuals from populations from the entire Balkan Peninsula are in progress.

**KEYWORDS:** Balkan Peninsula, *Fagus* sp., genetic structure, nuclear and chloroplast microsatellites

Poster presentation 53 02 64

### PRELIMINARY PHYLOGENETIC RESULTS OF MERWILLA (HYACINTHACEAE) BASED ON CHLOROPLAST DNA DATA

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The genus *Merwillia* Speta (Fam. Hyacinthaceae) was recently segregated from *Scilla* L. in a wide sense for some species endemic to the eastern parts of South Africa and southern Mozambique, differing from *Scilla* s. str., the latter being a genus endemic to the Mediterranean basin. Despite the popularity of *Merwillia natalensis* as a garden or medicinal plant, there is little information published on the taxonomy, systematics or evolution of *Merwillia*. This genus is usually considered as very complex based on its large morphological variability among populations, commonly occurring in the same geographical areas. Traditional taxonomic studies in *Merwillia* based on morphology accept four species: *M. natalensis*, *M. dracomantana*, *M. kraussii* and *M. lazulina*. However, there is still great confusion regarding the taxonomic limits in the group. Here, we aim to explore species boundaries and biogeographic patterns in *Merwillia* using molecular data based on cpDNA. We analysed a total of 29 *Merwillia* samples from 20 South African localities covering the whole distribution of the genus. The matK chloroplast gene was amplified. The main polymorphic parameters obtained by DnaSP had the following values: haplotype diversity (Hd) was 0.788, nucleotide diversity (Pi) was 0.00464, while the average number of nucleotide differences (K) was 4.158. Maximum Parsimony and Maximum likelihood trees were constructed. Both phylogenetic analyses have a similar topology revealing two clearly distinguished clades. Clades corresponded with geographical patterns of the *Merwillia* distribution, separating coastal and southern-lowland from the northern populations which include the populations at high elevation along the Drakensberg extending north and inland into Mpumalanga. Samples identified as *M. kraussii* were all located in the southern clade, but within this large clade some subclades were identified that included samples morphologically resembling *M. natalensis* and other forms.

Poster presentation 55 02 81

### ON THE ORIGIN OF SAFFRON: PHYLOGENY OF CROCUS SERIES CROCUS

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Saffron, *Crocus sativus*, is known as the most expensive spice by weight worldwide. *Crocus sativus* is a male-sterile triploid that is propagated vegetatively through its subterranean corms. To trace the evolution of triploid saffron, we performed a nested approach reaching from obtaining the phylogeny of the species group where saffron belongs to, through detection of its parents or closest relatives and identification of their distribution area. We clarified the relationships of the crop within series *Crocus* using five nuclear and chloroplast markers. We identified the diploid *C. cartwrightianus*, sometimes called 'wild saffron,' as the progenitor involved in the formation and domestication of saffron. Through the analysis of genome-wide distributed single-nucleotide polymorphism we were able to indicate the *C. cartwrightianus* populations in mainland Greece as probable progenitors of the crop. However, we could not find a *C. cartwrightianus* individual with the completely identical allele pattern of *C. sativus*, which is no surprise, as *C. cartwrightianus* is obligate outcrossing. Thus the allele composition of the individuals plants change between generations due to recombination.

**KEYWORDS:** Saffron, *Crocus sativus*, Origin

The Northern clade included several samples of *M. natalensis*, which split in different clades based on biogeography. Based on the large morphological variation of the genus and obtained phylogenetic clades, we suggest that ancient hybridization processes could have affected the current genetic diversity in the group. Moreover, the results points out to the necessity to recognize further species in the genus, although, further studies including additional molecular data based on chloroplast and nuclear DNA regions, are needed.

**KEYWORDS:** *Merwillia*, chloroplast DNA, matK gene, phylogenetic analyses

Poster presentation 54 02 60

### CORRELATION BETWEEN MORPHOLOGICAL FEATURES OF TRIFOLIUM ALPESTRE L. AND ELEVATION GRADIENTS IN BREZOVICA (SHARRI MOUNTAIN) – REPUBLIC OF KOSOVO\* (\*according to UNSC Resolution 1244)

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The object of research was *Trifolium alpestre* L. The area where the plant material of nine populations was collected was Brezovica (Sharri Mountain-Republic of Kosovo\*). The aim of research was to assess the ecological valence and distribution of this plant in Brezovica, impact of elevation gradients on some morphological features and morphological variability of these plant features between nine populations. The height of the plant, the length of flower calyx and corolla, the length, width and weight of the fruit were analyzed. The research showed morphological variability of plant features; the vertical distribution of *Trifolium alpestre* L. was very narrow.

**KEYWORDS:** *Trifolium alpestre* L., Brezovica, elevation gradients, morphological variability



### Session 3. Plant Molecular Biology and Genetics



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Poster presentation 01 03 06

#### MOLECULAR CHARACTERIZATION OF FIVE ENDEMIC BOSNIAN-HERZEGOVINIAN LAMIACEAE SPECIES

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The endemic plant species are important components of regional flora; they enrich the diversity and make flora of geographically close areas distinct. Limited distribution also makes them vulnerable. Therefore, the study of genetic structure of such species is an essential step towards understanding their evolutionary potential and finally planning conservation approach. Considering the long history of medicinal use of numerous Lamiaceae species in folk medicine, adequate conservation approach is necessary if sustainable use is to be achieved. The goal of the study was to obtain an insight into genetic diversity of five endemic Bosnian-Herzegovinian Lamiaceae taxa and explore their mutual relations. Specimens of five endemic Lamiaceae species (*Micromeria pulegium* (Rochel) Benth *Acinos orontius* (K.Maly) Šilić, *Satureja horvatii* Šilić, *S. subspicata* Bartl. ex Vis and *Thymus bracteosus* Vis ex Benth) were collected across their natural distribution area and morphologically characterized. The herbal material was dried and preserved in dark and dry area until further processing. The total DNA was extracted using modified CTAB based method. The specimens' cpDNA was analyzed by PCR-RFLP of *trnL* fragment and *matK* sequencing. Nuclear DNA was screened for polymorphisms by AFLP and sequencing of *ITS1* and *ITS2* regions. Statistical analysis including degree of population differentiation, molecular variance (AMOVA), Neighbor-Joining cluster analysis and geographic distance estimation was applied. Molecular characterization showed great diversity among five selected species. AFLP analysis showed comparable genetic diversity parameters for four species with *Micromeria pulegium* exhibiting lower degree of heterogeneity. *S. subspicata* samples form subgroups that correspond to the locations of sampling, which indicates possible link between genetic differentiation and geographical distance. With the exception of *S. subspicata* and *S. horvatii* where certain degree of genetic mixture is evident, three species are clearly genetically differentiated.

**KEYWORDS:** endemic Lamiaceae, AFLP, matK, nrDNA, ITS1, ITS2, DNA barcoding

Poster presentation 02 03 07

#### CYANOGENESIS CLINE OF WHITE CLOVER (*TRIFOLIUM REPENS*) POPULATIONS IN THE URBANIZATION GRADIENT OF THESSALONIKI (GREECE)

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As the human population keeps growing with accelerated pace, the urban environment has become a predominant feature of terrestrial landscapes. Urban environment – compared to rural – has very different environmental conditions that impose selection pressures to species; many of these species adapt through phenotypic and genotypic changes. *Trifolium repens*, commonly known as white clover, has a cosmopolitan distribution and the mendelian phenotypic trait of cyanogenesis. Cyanogenesis protects the plant against herbivores but in cold climates is selected against, because freezing temperatures lyse cells and trigger HCN release, causing self-toxicity in cyanogenic plants. It has been shown that *T. repens* evolves parallel clines in hydrogen cyanide HCN along urban-rural gradients in eastern North America. By studying the evolution of the HCN production in *T. repens*, the GLUE (Global Urban Evolution) project is an initiative that will provide the best replicated test of parallel evolution, on the largest scale ever attempted. Our research, part of the GLUE project, will test this hypothesis in Thessaloniki (N. Greece), a city with over one million inhabitants. We use the trait of cyanogenesis to observe the frequency that *T. repens* expresses it along a gradient from urban to suburban, periurban, and rural areas. To see if there is a cyanogenesis cline in the populations of *T. repens* along the urbanization gradient of Thessaloniki city, we collected 20 individuals from each of 46 populations following a transect of ca. 15 km from the urban center to the east rural areas. We conducted taxonomic identification of each individual we collected, to avoid sampling mistakes. Cyanogenesis was assessed for each individual using the Feigl–Anger assay. This work presents the preliminary results of the possible *T. repens* adaptation in Thessaloniki, the only city in Southeastern Europe and one of the few cities within the Mediterranean climate zone of the GLUE project.

**ACKNOWLEDGEMENTS:** The authors would like to thank the GLUE Project leading team for their support.

**KEYWORDS:** urbanization, adaptation, *T. repens*, cyanogenesis, Thessaloniki, GLUE

Poster presentation 03 03 13

**GENETIC DIVERSITY OF *POSIDONIA OCEANICA* MEADOWS IN THRACIAN (NORTHERN AEGEAN) SEA: PRELIMINARY RESULTS**

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*Posidonia oceanica* is an endemic seagrass widespread in the Mediterranean Sea, forming continuous meadows and playing an important role in coastal ecosystem dynamics. Up to date, it is considered the most important and well-studied seagrass of the Mediterranean. *P. oceanica* meadows of the Thracian Sea (Northern Aegean Sea, Greece) is estimated as one of the largest in the Eastern Mediterranean. Growing along the Mediterranean coastline, they are subject to continuous anthropogenic pressure, occasionally leading to decline, which challenges the ecological stability of those coastal ecosystems. Genetic diversity and intra-meadow heterogeneity in *P. oceanica* is correlated with adaptability of its populations in variable environmental conditions (both natural and anthropogenic) and therefore is an indicator of meadow's sensitivity to these perturbations. *P. oceanica* meadows showing signs of decline during 2000-2012, mainly in the easternmost part of the Thracian Sea, motivated us to estimate its genetic diversity using highly variable dinucleotide microsatellite markers. Samples were collected from eight stations in the Thracian Sea at a depth range of 9-11 m. Fresh leaf tissues, cleaned from epiphytes were grounded using distilled water and genomic DNA was extracted from eight samples widely distributed through the study area using an isoamyl-choroform protocol. Four microsatellite loci (Po5, Po5-10, Po5-49 and Po4-3) were amplified by Polymerase Chain Reaction and submitted to Sanger sequencing. Alleles were detected by manual and automated inspection (Mutation Surveyor software) of Sanger chromatograms in order to read superimposed base calls and identify the heterozygous alleles. Results were validated using 12% w/v polyacrylamide gel electrophoresis. Statistical analysis was conducted with appropriate software (GENCLONE v2.0, GeneAIEx, ARLEQUIN). Two loci were monomorphic (Po5, Po4-3) and two were polymorphic (Po5-10, Po5-49). Clonal diversity was found at 0.75 and levels of heterozygosity are exceeding the expected values. Our preliminary findings imply that the intra-meadow genotypic diversity is significant and provide an interesting insight concerning the correlation between genetic diversity and adaptability of *P. oceanica* in diverse environmental conditions.

**KEYWORDS:** *Posidonia oceanica*, marine angiosperm, intra-population genetic diversity, microsatellite

Poster presentation 04 03 05

**GENETIC DIVERSITY OF APRICOT CULTIVARS AND SELECTIONS FROM FACULTY OF AGRICULTURE, NOVI SAD BREEDING PROGRAM**

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Apricot selection program at the Faculty of Agriculture in Novi Sad, Serbia, started in early 80s with selection goals including late flowering, late frosts resistance, high productivity and good fruit quality. Selection from natural populations resulted with five cultivars and two selections still in the process of registration. Cultivars released from the selection program are nowadays wide spreading in Serbia and surrounding countries, making traditionally grown Magyar kajski and Kécskei rozsa less represented in the assortment of apricot cultivars grown in Serbia. Being selected from the limited cultivation area, although differing in phenotypes, their genotypic characterization has not yet been performed. The goal of the research was to determine the genetic diversity among apricot genotypes selected at the Faculty of Agriculture Novi Sad and compare them with 13 other cultivars grown worldwide. Plant material consists of 7 apricot cultivars and selection from Faculty of Agriculture, University of Novi Sad breeding program (Novosadska kasnocvetna, Novosadska rodna, NS 4, NS 6, Buda, SK1 and SK3), as well as 13 cultivars from other worldwide breeding programs. Young, fully developed leaves were collected, and total genomic DNA was extracted using modified CTAB protocol. A set of 17 SSR primer, from which 8 was associated with QTL-s for ripening time, PPV resistance, Xanthomonas resistance and chilling requirement according to GDR (Genome Database for Rosaceae) was used. High genetic diversity level was observed between all analyzed genotypes. Using described set of markers we were able to distinguish all genotypes with exception of SK 3, NS 4 and Magyar kajski. Cluster analysis grouped all cultivars from University of Novi Sad breeding program closely together while cultivar Buda was clustered away from this group. It can be concluded that all analyzed genotypes from Faculty of Agriculture, Novi Sad exhibited moderate level of genetic diversity assed by SSR markers. Regarding its phenotypic characteristics and genotype profile cultivar Buda is the most divergent from other genotypes arising from same breeding program.

**KEYWORDS:** breeding, diversity, molecular markers, SSR, microsatellites

Poster presentation 05 03 12

**MOLECULAR TOOLS FOR SUGAR BEET IMPROVEMENT IN INSTITUTE OF FIELDS AND VEGETABLE CROPS, NOVI SAD**

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Sugar beet is one of the most important industrial crops. Beet sugar now accounts for almost all sugar production in the Europe and for about 20% of total world production. Crop improvement is important in order to enhance productivity, sugar content or other desirable traits, and molecular techniques can help to accelerate such efforts. One of our research interests is focused on to the development of new sugar beet lines and hybrids combining marker assisted selection with conventional breeding methods. DNA markers were used to assess the genetic diversity and distance among breeding lines and correlate them with hybrid performance, in order to define the heterotic groups and predict heterosis effect. Our activities also involve the investigation of different reaction of sugar beet genotypes to reduced water supply, as the major limiting environmental factor in sugar beet growing. Genotypes with different ability to maintain turgor in the unfavorable field conditions were analyzed in the greenhouse and *in vitro* for morphological and physiological parameters of water regime under conditions of water deficit. The obtained plant material was also studied for the expression of genes that are known to respond to osmotic stress. There are reasons to believe that fulleranol ability to form hydrogen bonds with water molecules makes this nanoparticle a potential intracellular water depot, which can be used if osmotic stress occurs. In collaboration with colleagues from the Faculty of Science, Novi Sad we have started to analyze the influence of fulleranol on sugar beet plants exposed to drought stress. Results indicate that application of fulleranol can modify intracellular water metabolism and enable adaptation of plants to drought stress. The activities as part of phytopathological studies consider the main sugar beet leaf and root pathogens such as *Cercospora beticola* and *Macrophomina phaseolina*. The research presented here are an attempt toward fulfilling the potential of molecular tools in an improvement of sugar beet crop to better tolerate biotic and abiotic stresses.

**KEYWORDS:** crop improvement, molecular research, sugar beet

Poster presentation 06 03 11

**FUNCTIONAL ENRICHMENT ANALYSIS OF MARKER EFFECTS FROM GENOMIC PREDICTION MODELS IN SOYBEAN**

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Genomic selection is a promising tool that can increase the efficiency of crop breeding, being particularly useful for improvement of quantitative traits. This approach is based on prediction of plant performances by estimation of breeding values using simultaneously effects of molecular markers across the genome. Even though all available genome-wide markers are used for selection, it could be expected that markers located near genes responsible for particular biological function will have higher effects on final breeding value. This work aimed to estimate marker effects for soybean yield by developing prediction model and to perform functional annotation and enrichment in order to reveal the differences in marker effects related to different biological functions on yield prediction. Training population of 230 lines from the existing breeding program of Institute of Field and Vegetable Crops, was analysed using genotyping-by-sequencing and evaluated for yield in field trails over three growing seasons. Molecular, single nucleotide polymorphism (SNP) and phenotypic data were combined in prediction model, considering ridge-regression best linear unbiased prediction statistical method. Each SNP located inside predicted gene model was assigned with gene ontology (GO) terms for biological processes. The distribution of marker effects with particular GO term was compared with distribution of effects of all remaining markers by using Kolmogorov-Smirnov test. False discovery rate correction was performed, adjusting  $p < 0.05$  as significant. Functional enrichment identified 3471 unique GO terms, 1829 of which were related to biological processes. In total, it was identified 21333 SNPs associated with a biological processes GO terms, covering 10594 predicted gene models. GO terms were further clustered to 36 higher categories, including from 15 to 805 GO terms. Significant enrichment was observed for terms related to cell differentiation and cell death. Furthermore, cluster covering biosynthesis was also significant, including multiple terms related to protein, lipid, carbohydrate and nucleic acid metabolism, as well as secondary metabolism. This indicates that processes involving primary and secondary metabolites can be important for predicting yield breeding values. Preliminary data revealed associations with potential for elucidating biological processes relevant to yield, which should be deeply investigated.

**KEYWORDS:** soybean, genomic prediction, functional enrichment, gene ontology

Poster presentation 07 03 04

### INVESTIGATION OF ANTIMUTAGENIC EFFECT OF *VACCINIUM ARCTOSTAPHYLOS* L. ON MERISTEMATIC CELLS OF *ALLIUM CEPA* L.

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Antioxidants having neutralizing characteristics of free oxygen radicals being harmful for DNA molecules and causing cancer are mainly chemicals used to against the harmful effects of mutagen and carcinogen agents. Analysis of antimutagenic effects of antioxidants against these agents is very important for cell cycle. In this study the antimutagenic effect of anthocyanin-rich extract obtained from *Vaccinium arctostaphylos* L. growing naturally in Eastern Black Sea was investigated on meristematic cells of *Allium cepa* L. roots exposed to copper. Root tips of *Allium cepa* were first treated with 50 and 100 mg/L copper sulphate and then treated with anthocyanin rich extract for 12 and 24 hours. Mitotic index and various chromosomal aberrations were determined at the end of cytogenetic examinations. It was found that copper sulphate reduced the mitotic index in all experimental groups when compared with control groups and induced chromosomal abnormalities. Also, it was observed that anthocyanin treatment after copper sulphate treatment increased mitotic index in 50+ASY groups and reduced total abnormality rate. With the present study, the antimutagenic effect of *Vaccinium arctostaphylos*'s anthocyanin-rich extract against to copper concentrations was determined.

**KEYWORDS:** *Vaccinium arctostaphylos*, *Allium cepa*, copper, anthocyanin, antimutagen

Poster presentation 08 03 09

### IDENTIFICATION OF SOIL BACTERIA ASSOCIATED WITH METAL-TOLERANT *GENISTA PILOSA* L. GROWING IN SERPENTINE SOILS

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Serpentine (ultramafic) soils are characterized by low levels of essential nutrients and high levels of specific heavy metals

(such as nickel, iron, chromium and cobalt) thus representing very hostile environment for most plants. Hence, flora of these areas differs significantly from that of adjacent soils and includes mostly metal tolerant plants. Tolerance to heavy metals in plants is the result of genetic adaptation; however, rhizosphere bacteria were shown to possess properties that may influence plants' stress response. Interaction between selected bacteria and plants may improve the plants potential for metal accumulation and detoxification. The aim of our study was to isolate bacteria bound to rhizosphere of *Genista pilosa* L. growing in serpentine soil; to select metal tolerant colonies, identify them and cryopreserve interesting clones. Plant and soil samples were collected from serpentine soil near Olovo (Bosnia and Herzegovina) in May, 2017. Concentrations of heavy metals in soil and plant material were measured using atomic absorption spectroscopy (Shimadzu, 7000A). Rhizosphere bacteria were isolated from soil and root of *G. pilosa*. Based on differences in their morphology individual colonies were subcultured. Selection of metal tolerant strains was performed in tryptone yeast agar supplemented with metal salts (Cu, Cr, Ni, Co). Metal-tolerant clones were screened by sequencing of 16S rDNA. Sequencing was performed using Genetic analyzer 3500 (Applied Biosystems). Sequences were subjected to BLAST (Basic Local Alignment Search Tool) against NCBI (*National Center for Biotechnology Information*) database. Soil samples contain high concentrations of Fe, Ni and Co with low content of Ca. Concentrations of heavy metals in plant material do not exceed toxic levels. However, concentrations of Mn and Fe are quite high. Isolated rhizosphere bacteria showed resistance to Cu, Ni and Cr. Based on the obtained partial sequences of 16S rDNA and comparison with NCBI database entries bacteria isolated from soil samples showed highest sequence similarity to *Bacillus* genus while bacteria associated with root of *G. pilosa* showed 99% similarity to *Pseudomonas chlororaphis*. Future analyses will involve screening of plant growth promoting activity in the context of heavy metal tolerance.

**KEYWORDS:** heavy metals, serpentinophytes, rhizobacteria, metal tolerance, 16S rDNA

Poster presentation 10 03 03

### DETERMINING BORON RESISTANCE GENES IN BABY'S-BREATH (*GYPHOPHILA SPHAEROCEPHALA*) AND EUROPEAN ALKALI GRASS (*PUCCINELLIA DISTANS*)

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Boron (B) is an essential microelement for plant function. The range between optimum and toxic levels of boron for plant species is narrow; therefore, boron deficiency and tox-

icity have been commonly observed. Research showed the presence of plant species (*Gypsophila* spp., *Puccinellia distans*, *Elymus elongatus*) naturally growing in areas where the boron content is extremely high. In our study, to characterize genes conferring resistance to boron toxicity, baby's-breath (*Gypsophila sphaerocephala*) and European alkali grass (*Puccinellia distans*) were collected from a boron mining area in Turkey. *Gypsophila sphaerocephala* and *Puccinellia distans* plants were grown in hydroponic system containing 500 mg/L and 1250 mg/L boron, respectively. cDNA libraries were constructed from these two plant species, after exposure to high levels of boron stress. Using functional genomics approach, we aim to identify gene(s) in baby's-breath and European alkali grass which confer boron resistance. So far, candidate genes have been determined, and confirmation of them is in progress. Subsequently, intracellular localization and direct induction of mRNA levels in response to boron stress of the candidate proteins will be investigated by confocal microscopy and RT-qPCR techniques. The result of this research will help us to enlighten the molecular mechanisms of the candidate proteins in boron stress tolerance.

**KEY WORDS:** Boron toxicity, boron resistance, *Gypsophila sphaerocephala*, *Puccinellia distans*

Poster presentation 11 03 01

### GENOME-WIDE IDENTIFICATION AND EXPRESSION ANALYSIS OF O-ACETYL SERINE(THIOL)LYASE (OASTL) GENES UNDER CADMIUM STRESS IN SORGHUM (*SORGHUM BICOLOR* L.)

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Cysteine (Cys) is the first molecule in plant metabolism which includes both sulfur and nitrogen. It can be synthesized in three cellular compartments, containing chloroplast, cytoplasm, and mitochondrion. The final step of cysteine biosynthesis is catalyzed by the O-acetylserine(thiol)lyase enzyme. In the present study, seven members of OASTL gene family in sorghum (*Sorghum bicolor*) were identified at genome-wide scale and a comparative bioinformatics analyses were performed between sorghum and *Arabidopsis* OASTLs. In all OASTL proteins, pyridoxal-phosphate dependent domain structure (PALP, PF00291) was identified. The gene ontology annotations also revealed that all sorghum OASTL genes have KOG1252 (Cystathionine beta-synthase and related enzyme) and K01738 (cysteine synthase A) activities. In promoter sequences of OASTL genes, diverse cis-acting elements were found, including hormone and light responsiveness, abiotic stress responsiveness, and tissue-specific (meristem and

endosperm). Sorghum OASTL genes indicated medium or high levels expressions at anatomical parts and development stages based on digital expression data. Expression profiles of OASTL genes were also analyzed under cadmium (Cd) stress in sorghum by Real-Time quantitative PCR (RT-qPCR) and it was found that OASTL genes were mostly down-regulated. The predicted 3D structure of OASTLs indicated some structural diversities as well as variations in secondary structures.

**KEY WORDS:** cysteine, cadmium, sulfur, sorghum, co-expression, bioinformatics

Poster presentation 12 03 02

### DREB2 (DEHYDRATION-RESPONSIVE ELEMENT-BINDING PROTEIN 2) TYPE TRANSCRIPTION FACTOR IN SORGHUM (*SORGHUM BICOLOR* L.): BIOINFORMATICS AND EXPRESSION ANALYSES

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The dehydration responsive element binding proteins (*DREB*) are important transcription factors (TFs) that are induced by abiotic and biotic stresses. They can be categorized by two subgroups: *DREB1* and *DREB2*. In this study, we first performed bioinformatics analyses of *DREB2* genes. Later, expressions of *DREB2* genes were evaluated under cadmium (Cd) and salt (NaCl) stresses in sorghum (*Sorghum bicolor* L.). For bioinformatics analyses, gene/protein sequences were analyzed using bioinformatics tools. The expression patterns of *DREB2* genes were performed in roots and leaf tissues using RT-qPCR. Based on genome-wide analyses *DREB* genes in sorghum genome, the six putative *DREB2* genes were identified and all genes contained AP2 domain (PF00847) structure. The nucleotide variations of *DREB2* genes were found as  $\pi$ : 0.53 and  $\theta$ : 0.39, respectively. The phylogenetic analyses revealed a monocot-dicot divergence in phylogenetic tree. The 56 cis-regulatory elements were determined in promoter regions of *DREB2* genes, such as light responsive, hormone responsive, stress responsiveness, and tissue specific. According to digital expression analyses, *DREB2A* and *DREB2B* genes exhibited the more dynamic regulations in cell metabolisms. In NaCl and Cd stresses, *DREB2* genes showed various expression profiles and all *DREB2* genes in roots were up-regulated under salt stress. In Cd stress, *DREB2D* gene was particularly up-regulated in leaves and roots. In addition, the predicted 3D structure of *DREB2* proteins showed the some structural divergences. In conclusion, our findings could contribute understanding functions of *DREB2* genes in sorghum under abiotic stress conditions and could be scientific base for crop improvement studies, particularly in sorghum.

**KEYWORDS:** *Sorghum*, abiotic stress, bioinformatics, gene expression, *DREB2* gene

## Session 4. Floristics, Vegetation and Phytogeography



Balkan  
Botanical  
Congress

Introductory lecture 01 04 57

### BALKAN ENDEMIC PLANTS IN THREE BALKAN COUNTRIES - SIMILARITIES VERSUS SPECIFICITIES

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Endemic plants represent unique characteristics of the Balkan peninsula vascular flora. During last decade, the analyses of the Balkan endemic species and subspecies within Serbia, Bosnia and Herzegovina and Montenegro were accomplished. The aim of this work is to make comparative analysis of the Balkan endemics in three Balkan countries in order to emphasise similarities and specificities within the studied areas. Overall, there are 749 endemic taxa that grow in these countries, of which 506 are present in Serbia, 378 in Montenegro and 312 in Bosnia and Herzegovina. Similarity analysis shows that 124 taxa are common for all three countries, while 208 plants are present in Serbia and Montenegro, 215 plants are common for Montenegro and Bosnia and Herzegovina and only 148 taxa are distributed both in Serbia and Bosnia and Herzegovina. Contrary to the similarities, there are 275 Balkan endemic species and subspecies that are recorded only in Serbia, 78 only in Montenegro and 73 only in Bosnia and Herzegovina. Jaccard's Indexes of Similarity of the Balkan endemic flora between pairs of countries are relatively low: Serbia and Montenegro (30.8%), Serbia and Bosnia and Herzegovina (22.1%) and Montenegro and Bosnia and Herzegovina (45.3%). In all three countries, Asteraceae, Caryophyllaceae and Fabaceae are represented with the highest number of endemic taxa, while the richest genera are *Hieracium* and *Dianthus*. South European mountain and Central European mountain chorological groups are the most dominant in the Balkan endemic flora of three countries. Most endemics were recorded on limestone in all three countries, while in Serbia and Bosnia and Herzegovina, there are certain number of endemics that inhabits ultramafic substrates; silicate endemic flora is well represented only in Serbia. High-mountain areas of the central Dinarides are the most important centres of floristic diversity in Bosnia and Herzegovina (Mts Maglić, Volujak, Prenj, Čvrsnica, Čabulja), while in Montenegro the highest number of endemic taxa was recorded in Mts Durmitor, Komovi and Lovćen. In Serbia, Mts Prokletije, Šar Planina, Paštrik and Koritnik are characterized by the highest level of endemism, while Mts Kopaonik and Stara Planina are distinctive centres of floristic richness of the silicate areas.

**KEYWORDS:** flora, endemics, Balkan Peninsula, Bosnia and Herzegovina, Montenegro, Serbia

Introductory lecture 02 04 79

### MORE THAN AN APPENDIX – POPULATIONS OF INNER-ALPINE AND WESTERN BALKAN STEPPE SPECIES ARE MORE DIVERGENT AND CONSERVATION RELEVANT THAN ANTICIPATED

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Extrazonal steppes include those in inner-Alpine, continental valleys and adjacent areas, for instance the western Balkan Peninsula, and cover a relatively small area. In contrast, the zonal Eurasian steppes are huge, spanning from the Pannonian Basin to eastern Central Asia. The results of a recently completed project clearly reject the hypothesis that steppe organisms generally reached the Alps and the western Balkan Peninsula by recent range expansion from east to west during cold periods of the late Pleistocene. Rather, our results emphasise that, from a biodiversity point of view, extrazonal steppes are not the mere appendix of the zonal Eurasian steppes. In more detail, we showed that in five out of six investigated species, populations from extrazonal steppes are strongly divergent from those from zonal steppes. The divergence is partly so pronounced that partly even descriptions of new, European endemic taxa are required. Our data also reject the hypothesis that the phylogeographic patterns of steppe plants and animals are congruent, which implies idiosyncratic responses of species to climatic oscillations instead of range shifts of entire communities. Finally, for some of the investigated species, the fine-scale resolution facilitated by genome-scanning data provided evidence for genetic exchange among insular steppe habitats of different valleys implying that high mountain ranges do not pose barriers to dispersal. The fact that in several species extra-zonal populations of steppe biota need to be treated as separate evolutionary entities, independent from their zonal distribution, strongly emphasises the conservation value and the uniqueness of the remaining dry grasslands in the Alps and adjacent areas. We are confident that our findings will act as a new basis for conservationists and policy makers to advocate a future, scientifically founded conservation of extrazonal steppes in Europe.

**KEYWORDS:** steppes, West Balkan, phylogeography

Introductory lecture 03 04 47

## BALKAN DRY GRASSLAND DATABASE - OVERVIEW, CURRENT STATUS AND FUTURE PERSPECTIVES

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The Balkan Dry Grassland Database (BDGD) was established in 2012 for studying the habitat and floristic diversity of dry grasslands in the region. It is registered in GIVD (EU-00-013) and is a part of the European Vegetation Archive (EVA) and the sPlot database. The BDGD comprises data of various dry grassland classes. It also includes data of neighbouring classes such as *Molinio-Arrhenatheretea* (order *Arrhenatheretalia*), *Elyno-Seslerietea*, *Caricetea curvulae*, *Daphno-Festucetea*, *Trifolio-Geranietea* and *Nardetea strictae*. To date, BDGD contains 9,852 relevés, which are stored in Turboveg v2.101 software. The species list is based on the one of the Bulgar-

ian National Database, which was implemented with hundreds of new taxa from the region. The majority of the data in the BDGD was digitized from published literature sources (78.2%), while the rest are unpublished relevés. The BDGD consists of data from 8 countries. Geographic coordinates are available for 62.8% of the relevés, most sources being geo-referenced *a posteriori*. Plot size ranges from 1 to 4000 m<sup>2</sup> (the most frequently used plot sizes are between 15 and 100 m<sup>2</sup>). Total vegetation cover is provided for 77.8% of the relevés. Approximately 83% of the relevés in the BDGD are classified into syntaxa of different levels. The BDGD is a good starting point for various functional diversity related research questions. This database will be useful for a wide range of researchers, the nature conservation administration and non-governmental organisations in the region.

**KEYWORDS:** dry grasslands, Balkan peninsula, vegetation databases, TURBOVEG, ecoinformatic

Introductory lecture 04 04 78

## PHYTOGEOGRAPHY OF GREY ALDER (*ALNUS INCANA*) PHYTOCENOSES IN THE DINARIDES

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Grey alder (*Alnus incana* (L.) Moench) is an European species distributed across central, northern and eastern Europe up to the Caucasus. Contrary to its more widespread congener, the typical lowland common alder (*Alnus glutinosa* (L.) Gaertn.), grey alder is more common in higher elevations, up to 1400 m, where it mostly forms stands alongside alpine rivers, brooks and other humid habitats. Its communities were recently studied on the European level from the point of phytosociology, but questions about phytosociological diversity, biogeography and relationships to other parts of Europe remained open, since the whole Balkan area was underrepresented in this study. Namely, most of previously published data were written in national languages and published in hardly accessible local journals, representing "grey information", often neglected in modern studies. Therefore, the aim of our study was to use our recent field researches and investigate the published data more thoroughly, to fill this gap. Stands from the south-eastern Dinarides were analysed and compared with the ones from the north-western Dinarides. The comparison revealed significant differences in the floristic composition and the degree of differentiation, particularly regarding the presence of the Illyrian floristic geoelement. In total 198 relevés were analysed based on floristic composition and grouped in four main clusters, out of which three are confined to the north-western Dinarides, showing that this is the centre of diversity of Dinaric grey alder phytocenoses. For each cluster, characteristic and differential species were estimated, clearly defining the ecological and phytogeographical differentiation of these phytocenoses. However, further research is inevitable in order to gain a more detailed picture of the characteristics, diversity and relationships between Dinaric stands of grey alder.

**KEYWORDS:** floristic geoelements, forests, phytosociology, vegetation ecology

Oral presentation 05 04 12

## MAPPING THE FLORA OF GREECE

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The recent publication of a *Checklist of the Vascular Plants of Greece* (2013, 2016) as well as *Atlas of the Aegean Flora* (2016) mark significant progress for plant taxonomy and phytogeography in Greece. The author is now working on an *Atlas of the Greek Flora* which is planned to contain detailed distribution maps for all species and subspecies of vascular plants in the country (c. 6,600). Maps are being generated from the *Flora Hellenica Database* which was started in 1989 and now contains a little over one million geo-referenced records. Practically all published records (from c. 2,600 publications) have been critically registered for the database; in addition, significant parts of the holdings in the most important public herbaria (ATH, B, C, G, LD, UPA, W, WU, etc.) have been included. 478,877 database records refer to herbarium specimens, 311,248 to published floristic reports, and the rest (c. 230,000) to field notes, photographs, seed collections, etc. 410,109 database records have been registered by the author, 139,316 by Britt Snogerup (Lund), and the rest by c. 45 operators. As far as possible, herbarium specimens and literature reports have been critically evaluated, and great care has been taken in data validation to minimize the number of errors. The basic structure of the database has remained almost unchanged since the start in 1989, but the technical platform has been continuously updated. The *Atlas of the Greek Flora* will serve as a general reference work and as a basis for further research into Greek plant biodiversity and biogeography. Soon after publication the whole dataset will be made available electronically to *bona fide* researchers on the Greek flora. In the oral presentation a number of examples are being shown of distribution types, biogeographical elements, etc. The *Flora Hellenica Database* will be a powerful tool for further analysis of biogeographical patterns.

**KEYWORDS:** flora, Greece, phytogeography, databasing

Oral presentation 06 04 04

## THE FLORA OF AMORGOS

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Amorgos island is situated at the eastern margin of the Kiklades in the South Aegean Sea, ca. 220 km southeast of Athens. The island has an area of 121 sq km, excluding the small adjacent islets. It is dominated by three mountain ridges

aligned northeast to southwest; these are of great importance, affecting the local climate and thus the vegetation. Although possibly the driest island in the Kiklades with an average rainfall of 350 mm, approximately 1100 taxa have been recorded from the island. The central Aegean flora is relatively poor in unique elements but at least 28 Greek endemics occur which is noteworthy considering the island's small size. These endemics are illustrated together with their distribution maps. The maps are based on our own records and from verified reports. Fieldwork has been carried out in all seasons, so the autumn, winter and vernal flora of the island is rather thoroughly documented. This provides valuable information on annuals, spring and autumn geophytes. A vegetation map is established based on registered plant records and field notes. The phytogeographical affinities between Amorgos and six other Greek islands are calculated. There are several species which do not occur in the Kiklades or central Aegean but they were found on Amorgos. Rechinger had noted the absence of several taxa from the central Aegean area and termed the apparent floristic gap the "Kykladen-Fenster".

**KEYWORDS:** Kiklades (Central Aegean area), Greece, flora, vegetation, endemics, distribution maps

Oral presentation 07 04 08

### GLACIAL RELICTS IN THE MEDITERRANEAN DINARIDES – A PHENOMENON OF COLD-AIR POOL MICROCLIMATES?

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Glacial relict species and arcto-alpine vegetation are reported from an isolated Mediterranean enclave in the Opuvani do (1575 m a.s.l.) cold-air pool (CAP). Previous research assumed that glacial relict/refugia flora and boreo-alpine phytochorions are absent in the maritime High-Karst physiographic area in the Mount Orjen Range in Montenegro. The recent detection of residual boreo-alpine biocoenoses (*Salicion retusae*, *Scabioso silenifoliae-Dryadetum octopetalae*, *Drepanoclado uncinati-Heliospermetum pusillum* and *Allietum schoenoprasi*), 70 km south of known Dinaric inland localities, calls this assumption into question. The investigated glacially transformed paleodolines are located below the timber line. Cryophilous species and snowbed ecotypes inside the closed depressions arrange vertical with bottom-up stratification regulated by freezing tolerances leading to vegetation inversion. Local topographic (elevation, depth and sky-view) and terrain parameters (geology and geomorphology) affect the magnitude of diurnal cold-air drainage. The persistence of the glacial relict flora is mainly related to ecogeomorphic feedbacks from plant-soil processes in periglacial environ-

ments (e.g. cryopedogenetic phenomena associated to soil downslope creeping, frost heave and rock surface crushing). Long snowpack duration, periglacial weathered debris topplings and lower temperatures influence soil moistures, leading to late depletion of moisture pools. Ecological surrogates (e.g. *Dryas octopetala*, *Salix retusa*) offer valuable information on species distributions dependent on topographic and ecogeographic factors. The surrogates were tested as indicators for periglacial microclimates, severity of geomorphic disturbances (i.e. solifluction) and prolonged snow activity. Dinaric paleodolines provide essential conditions for long-term resilience of oro-arctic phytochorions (calcareous snow-beds, arcto-alpine heath and boreo-montane peaty meadows) during previous and present warm phases. Potential deviation of climatically suitable alpine habitats or severe changes in ambient ecological conditions is buffered relative to the surrounding areas (summits and ridge tops), which are not decoupled from the regional climate of the free atmosphere. Effects of topoclimatic complexity in the investigated CAPs showed substantial influence on landscape dynamics and persistence of glacial relict species.

**KEYWORDS:** cold-air pools, Dinaric glaciokarst, *Dryas octopetala*, solifluction, glacial relicts, ecogeographic analyses

Oral presentation 08 04 25

### VASCULAR PLANT DIVERSITY ON THE CHRISTIANA ISLAND GROUP AND ASPRONISI ISLAND (CYCLADES, GREECE)

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The Christiana Island group is the oldest part of the broader volcanic Santorini Archipelago and consists of three rocky islands, viz. Christiani, Askania and Eschati (the latter bare of vascular plants), located 23 km southwest of the Santorini island. Up to now, four attempts have been made to record the islets' vascular flora, with the earlier ones taking place in 1960 and 1967, followed by two recent attempts conducted in April 2017 and April 2018. Cumulatively, 171 taxa have been found in the Christiana island group, i.e. 159 taxa in Christiani and 89 taxa in Askania. Both islands share 77 taxa. 81 taxa have exclusively been recorded in Christiani but not in Askania, 13 taxa have exclusively been recorded in Askania but not in Christiani. From the total number of the 171 taxa, *Anchusa stylosa* subsp. *spruneri*, *Cotula coronopifolia*, *Eryngium amorginum* and *Fagonia cretica* are of major floristic interest. Aspronisi (0.1 km<sup>2</sup>) is the smallest of the ring islands surrounding the seawater-filled caldera (formed by the Minoan eruption of the Santorini volcano in 1645 BC). The uninhabit-

ed island was floristically unexplored prior to May 1990, when 46 taxa were collected on the island with 11 shore taxa added in April 2018. Beyond presenting the current knowledge of the vascular plant diversity, our study aims at additional analyses of the chorological and life-form spectra of the islets, indicating the predominance of Mediterranean species and therophytes, respectively. Moreover, we analyze and compare the taxonomic and functional diversity among the individual islands of the Santorini Archipelago. For this purpose, we calculated species richness and functional richness indices, using data for 26 functional traits for the 171 taxa. The values of these indices, as well as the values of each functional trait were compared among the islands with their different surface area and geological age in order to examine the species-area relationship and the species-time relationship, respectively. Both species- and functional- richness increase with increased area and geological age. According to theoretical expectations, the results of the individual traits analysis indicate a differentiation in plant functional characteristics between early and late stages of succession.

**KEYWORDS:** Christiana, Aspronisi, Santorini Archipelago, Aegean islands, flora, diversity, functional traits

Oral presentation 09 04 44

### DISTRIBUTION AND CONSERVATION STATUS OF THE ENDEMIC PLANT TAXA IN ALBANIA

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With ca. 3650 plant species in a small area of 28,748 km<sup>2</sup>, floristically Albania is ranked among the richest countries of the Europe. The recent data on plant species in Albania, published during this decade, allow us to re-evaluate the current knowledge on the state, distribution of endemic plants and their hotspot areas, altitudes and corresponding vegetation zones. Based on our data collected along 15 years and the literature, in Albania there are 68 endemic taxa or 54 species and 14 subspecies that belong to 19 families and 40 genera. The data show also that the southern floristic region is richer with 45 taxa, compared with 23 taxa found in the north and north-eastern part of the country. With respect to species distribution, Tomorri Mountain and Acroceraunian Mountains range have remarkably high presence of endemic species. Generally, most of endemic taxa occur in the altitudes from 400 m up to 1500 m, on open grasslands in between shrubs and forests of the Sub-Mediterranean and Sub-Alpine regions. Regarding conservation status, from the total number of endemics, only 12 species are considered as threatened one, where six species are assessed as CR, two species as EN and four species as VU. The high number of species and the low

numbers of endemics, compared to neighbouring countries such as Greece with 913 species, Italy with 712 species and Macedonia with 135 species, indicate that the Albania should have larger number of endemic plant taxa as far as we know it at present.

**KEYWORDS:** Albania, endemic taxa, threatened, Tomorri Mt and Acroceraunian Mts

Oral presentation 10 04 26

### TAXONOMIC DIVERSITY PATTERNS IN THE NORTHERN SPORADES ISLETS COMPLEX (GREECE)

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The Northern Sporades is a group of 24 islands and islets, located along the eastern coastline of mainland Greece, north-east of Evvia island in the Aegean Sea. This island group is designated as a NATURA 2000 protected area and forms a part of the „National Marine Park of Alonissos and Northern Sporades“. Herein, we focused on 20 of the 24 islets and islands; their area, elevation and distance from the nearest mainland ranges from 0.0038km<sup>2</sup> to 24.7595km<sup>2</sup>, 1 to 570m and 50.9 to 78.4km, respectively. The Northern Sporades are continental in origin, as most of the Aegean islands, and are dominated by calcareous-alkaline rocks. Our main aim is to fill the gap of floristic knowledge of the total vascular plant species richness of the Northern Sporades; hence, the subsequent interrelated objectives are a) to assess the taxonomic alpha- and beta- diversity patterns of the Northern Sporades islets complex via Generalized Dissimilarity Modelling and b) to investigate the phytogeographical relationships among the studied islets by applying a hierarchical clustering approach. A total of 729 plant taxa, (41 of which are Greek endemics) belonging to 355 genera and 91 families have been recorded in the study area, reflecting rather well the geographical and the bioclimatic characteristics. Therophytes and the Mediterranean chorological element predominate, as expected. Eight local endemics, two of which are single island endemics occurring in the study area, represent the West Aegean endemic chorological element. Nine islet specialists – species preferring isolated islets and cliffs unaffected by human activities – occur in 19 islets of the study area. The most species-rich families of the Northern Sporades are *Asteraceae*, *Fabaceae* and *Poaceae* summing up to 34.9% of the regional vascular-plant diversity. The alpha-diversity is rather high and high-

lights the important role of these islets with respect to the conservation of the plant diversity in the Aegean archipelago. The calculation of the floristic cross-correlations between the islets, by means of Sørensen's index, revealed very low floristic similarity even between neighboring islets. Regarding the beta-diversity patterns, environmental filtering and not dispersal limitation seems to shape the plant assemblages of the Northern Sporades islets.

**KEYWORDS:** Aegean, beta-diversity, biogeography, endemism, environmental filtering

Oral presentation 11 04 81

### GEOGRAPHICAL DISTRIBUTION OF DIPLOID AND TETRAPLOID POPULATIONS OF *JUNIPERUS SABINA* L.

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Being uncommon in Conifers, polyploidy was noticed in *Juniperus* genus. It has been reported in *Juniperus chinensis*, *Juniperus squamata*, *Juniperus virginiana* and *Juniperus thurifera*. Nowadays, only one population of *Juniperus sabina* has been found to be tetraploid in the Dinaric Alps. Interestingly, this population was examined recently using chloroplast markers, and classified as a distinct variety; *J. sabina* var *balkanensis*. This variety has been recently shown to be distributed in the Balkans, Italy and the western edge of Turkey. *Juniperus sabina* var *balkanensis* was found to combine *J. sabina sensu stricto* nuclear DNA and *J. thurifera* chloroplast DNA revealing a chloroplast capture event from an ancestor of *J. thurifera* lineage. This result witnesses the occurrence of an ancient hybridization event between these two species. The aim of this study is to assess the geographical distribution of the ploidy levels among *J. sabina* populations with a focus in the Balkan region. Genome size measurement was performed by flow cytometry on dried leaves of 18 populations of *Juniperus sabina* var. *sabina* and *J. sabina* var. *balkanensis*. The results showed that all studied populations of *J. sabina* s. s. were diploid (~20 pg/2C). However, all *J. sabina* var *balkanensis* populations were tetraploid (~40 pg/2C). Further genome size measurements are currently being processed for Bulgarian populations in order to extend the geographical area of the survey. These results open new opportunities for future research concerning the role of polyploidization and interspecific hybridization in the evolution of *J. sabina*.

**KEYWORDS:** polyploidy, genome size, *Juniperus sabina*, *Juniperus thurifera*, evolution

Oral presentation 12 04 01

### GRASSES (POACEAE) OF VOJVODINA (N SERBIA), RESULTS OF HERBARIUM REVISION

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Grasses are often neglected by field botanists. But their flora is rich and in temperate regions they represent about 1/10 of regional vascular plant floras. For the region of Vojvodina (N Serbia) around 200 species of grasses is expected. The aim of the study was to put in order the material, identify also the non-determined vouchers, produce distribution maps as result of the revision and prepare a determination key for publication. During revision of herbarium material of grasses in two main herbaria for the region (BUNS, PZZP) about 6000 sheets were analysed. Only the genus *Festuca* (s. lat.) has been left aside. Some common species were collected all-over the region, few dozen forest taxa distinctly linked only to the areas of Fruška gora and Vršачki breg, some taxa of high nature conservation importance to the areas of Deliblatska and Subotičko-Horgoška peščara. Some very interesting rare taxa were recorded in the preserved lowland marshes as e.g. *Agrostis canina*, *Calamagrostis canescens* and *C. pseudophragmites*. Some critical groups that need further research are in the genera *Puccinellia*, *Bromopsis*, *Koeleria* and *Stipa*. In the later quantity of available material and its quality was simply not sufficient for clear-cut recognition of taxa, but at least 5 species are here. Some new or rarely reported taxa were found partly in studied herbarium material, partly by late-autumn field excursions. Some represent first records for the studied territory, some are just confirmation of already known localities, some demand further study: *Aira elegans* subsp. *elegans*, *Alopecurus myosuroides*, *Arundo donax*, *Avenella flexuosa*, *Bromopsis ramosa*, *Calamagrostis canescens*, *C. pseudophragmites*, *Ceratochloa* sp., *Cynosurus echinatus*, *Gaudinia fragilis*, *Glyceria declinata*, *Hordeum murinum* subsp. *leporinum*, *H. secalinum*, *Koeleria pyramidata*, *Miscanthus sinensis*, *Molinia arundinacea*, *Panicum dichotomiflorum*, *P. miliaceum*, *Setaria faberi*, *S. italica*, *Sporobolus indicus*, *Stipa joannis*, *Taenatherum caput-medusae*. Among the most active collectors in both herbaria were V. Budak, R. Perić, M. Obradović, P. Boža, B. Butorac and V. Stojšić. Database that resulted from revision showed us also the geographical gaps where more systematic sampling is needed. Obviously grass flora outside the nature-protection areas (that include also the mentioned localities) is not known well.

**KEYWORDS:** Poaceae, Gramineae, flora, herbarium revision, Vojvodina

Oral presentation 13 04 63

### GRASSLAND VEGETATION OF SERBIA-CLASSIFICATION, ECOLOGY AND BIODIVERSITY

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Grassland vegetation is of great importance for livestock production in Serbia. The large data set of grasslands classified into *Molinio-Arrhenatheretea* and *Festuco-Brometea* classes in Serbia were analyzed to produce the overview of its classification, environmental conditions and biodiversity patterns. Phytocoenological relevés from relevant literature sources and our own investigations were stored in the Vegetation Database of Serbia (EU-RS-002). The final dataset contained 3346 relevés and 1553 species. Species composition was classified hierarchically by the beta flexible method and species ecological indicator values were used for the estimation of the ecological conditions. We identified 23 clusters, which were well grouped into five groups: A- mesic grasslands from *Molinio-Arrhenatheretea* class, B- Balkan dry grasslands from *Festucion valesiatae* and *Chrysopogono-Danthonion alpinae* alliances, C- steppe and sand grasslands from *Festucion rupicola* and *Festucion vaginatae* alliances, D- Balkan submediterranean grasslands from *Scabioso-Trifolion dalmaticae* alliance and E- Balkan ultramafic rocky grasslands from *Halacsyetalia sendtneri* order. The classification problems, environmental conditions and biodiversity patterns of investigated grassland vegetation were discussed. The high floristic diversity and conservation relevance, with large number of Balkan endemic species (204), was exhibited by *Festuco-Brometea* class.

**KEYWORDS:** Grassland vegetation, *Molinio-Arrhenatheretea*, *Festuco-Brometea*, Serbia

Oral presentation 14 04 30

### CHRYSOPOGON GRYLLUS IN LOESS GRASSLANDS ON THE SOUTHERN LIMITS OF THE CARPATHIAN BASIN

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Scented grass, *Chrysopogon gryllus* (L.) Trin, has wide ecological amplitude and appears in many floristically and ecologically different grassland communities across its distribution range. We studied dry grasslands dominated by *Chrysopogon gryllus* on loess substrate in the transitional zone between the Pannonian and Mediterranean region of the Balkan. *Chrysopogon gryllus* dominated communities are considered to be the most valuable grassland type here, comprising species with different chorological characteristics (most interesting are e.g. pannonic, continental and sub-Mediterranean floristic elements). Our goal was to determine what kind of patterns can be observed after abandonment of grazing in these grasslands. Temporal changes of the cover values of dominant grasses and patterns of floristic richness were analyzed upon coenological relevés which were recorded in permanent plots between 1999 and 2016. Data collected in abandoned grasslands were compared to data from other grassland types. Our results were in accordance with theoretical expectations and previous preliminary results, which showed great variations among sample sites. The cessation of grazing and burning in semi-natural dry grasslands with *Chrysopogon gryllus* has caused a further increase of its cover from 20% to 60%. This species appeared to be a stronger competitor than *Botryochloa ischaemum* and *Brachypodium pinnatum*, whose cover decreased proportionally in the same permanent plots. In the mid-successional stage of previously grazed and abandoned old fields, the cover of dominant *Poa angustifolia* decreased (from 35 to 5%), while *Botryochloa ischaemum* cover fluctuated and slightly increased (from 10 to 30%). Abandonment of semi-natural grasslands with *Chrysopogon gryllus* has resulted in rapid exchange of dominant species and at the same time showed a drastic decrease of floristic richness. High dominance of scented grass and poor floristic richness was also observed in the pastures abandoned for decades after severe overgrazing. The results of our long-term studies of vegetation dynamics in grasslands can contribute to the scientific foundation of their biodiversity conservation and optimal grassland management.

**KEYWORDS:** abandonment, floristic richness, loess, permanent plot, trade-off, vegetation cover.

Oral presentation 15 04 65

## PONTIC-PANNONIAN SAND VEGETATION – SYNTHETIC OVERVIEW

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This paper analyses and synthesizes the present knowledge of dry grassland vegetation on sandy soils in South-Eastern Europe and surrounding areas. This vegetation is of high natural value both at European and national scales and include communities dominated by perennial tussock-grasses and herbs, with frequent annuals and cryptogams, typical of nutrient-poor, mostly base-rich, sandy soils and mostly appearing in the continental climate, with cold winters, often with long frosts and shallow snow, and hot, droughty summers. According EuroVeg Checklist this vegetation belongs to class *Koelerio-Corynephoretea canescentis* Klika in Klika et Novák 1941. For our analysis, we used a dataset of 95.415 plots and 21.471 species, obtained from European Vegetation Archive (EVA). Selection of data was performed by applying a set of different criteria. After removal of duplicates, harmonization and verification of taxonomy and nomenclature, removal of plots dominated by shrubs and trees, the final database was formed. The analyses have been carried out on 2.966 relevés and 1.667 species. We perform hierarchical cluster analysis with Pc Ord, where the species cover data were transformed to presence or absence of species (due to the problem of transforming the cover values of different scales), using the distance measure by Sørensen (Bray-Curtis), and Flexible Beta (-0.25) as a linkage method. Crispness of the classification is tested and local maxima of crispness was at 2 and 6 clusters. In the first division of the data set, a clear difference between the two groups emerged. This dissimilarity points to distinction between subatlantic inland sand dune vegetation and (sub)continental fescue sandy steppes in the foreststeppe and steppe zones of Europe, due to differentiation of macroclimate and differences in the substrate. This two groups differ in their floristic, ecological and geographical characteristics. The next classification level implies a clear grouping of relevés into 6 clusters. For each of these six clusters the area of distribution and diagnostic plant species were determined. Dissimilarity is based on different stages of vegetation successions and ecological differentiation of site conditions.

**KEYWORDS:** sand vegetation, hierarchical cluster analysis

Oral presentation 16 04 14

## PHYTOSOCIOLOGICAL ACHIEVEMENTS AND GAPS IN THE KNOWLEDGE OF THE SUBMEDITERRANEAN THERMOPHILOUS OAK FORESTS AND WOODS IN BULGARIA

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The submediterranean thermophilous oak forests and related to them woods are widespread and zonal vegetation in Bulgaria. The aim of this study is to increase the level of knowledge on their classification as well as the phytogeographical and ecological gradients in their floristic composition and ecology. Phytosociological classification is proposed on the basis of 725 relevés from all regions of Bulgaria. Classical numerical analysis was done using some special programs (SYNTAXA, JUICE, and CANOCO) applying the classical Braun-Blanquet's approach. The results are classification scheme of the most widespread oak forests and woods in Bulgaria, including the communities dominated by *Quercus cerris*, *Q. frainetto*, *Q. pubescens*, *Q. robur* ssp. *pedunculiflora*, *Q. petraea* gr., *Q. hartwissiana* and *Q. coccifera*. All main alliances of the thermophilous deciduous oak forests and woods were established in Bulgaria: *Aceri tatarici-Quercion confertae*, *Quercion petraeo-cerridis*, *Carpinion orientalis* and the Euxinian alliance *Carpino orientalis-Acerion hyrcani*. Some peculiarities of their distribution related to the biogeographical influences, climate, soils, altitudes, etc. are emphasized and presented. The thermophilous oak woods and forests in Bulgaria have specific and rich diversity which follows the gradient between typical continental influence on north, to the sub-Mediterranean dry to the south-west and Euxinian humid to the south-east direction. But it also depends on local conditions such as elevation, exposition, soil richness and humidity, etc.

**KEYWORDS:** syntaxonomy, numerical analysis, xerothermic forests

Oral presentation 17 04 05

## CHASMOPHYTIC VEGETATION OF THE CENTAUREO CUSPIDATAE-PORTENSCHLAGIELLION RAMOSISSIMAE (SOUTH-EASTERN ADRIATIC, NE MEDITERRANEAN)

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The *Centaureo-Portenschlagiellion* describes the cliff vegetation of the south-eastern Adriatic. This habitat type harbours many endemic taxa and has a high value for biodiversity conservation. Notwithstanding its importance, knowledge about the syntaxonomy of this alliance is still poor. This paper aims at revising the synchorological, coenological and floristic relationships of the associations of the *Centaureo-Portenschlagiellion*. The revision is based on a data set of 103 relevés of Mediterranean xerothermic cliff from Croatia, Bosnia and Herzegovina, and Montenegro. The relevés were clustered by using the Flexible beta method. The Indicator Species Analysis was used to identify the diagnostic taxa of the main clusters of relevés and NMDS ordination was undertaken to visualize the floristic relationships among them. Results revealed that only seven associations belong to the *Centaureo-Portenschlagiellion* whereas the others already described in phytosociological literature, were invalidly described or should be treated as syntaxonomic synonyms of the previous ones. Two associations (*Inulo-Centaureetum cuspidatae* and *Portenschlagiello-Campanuletum portenschlagianae*) were reduced to the rank of subassociations of the *Moltkio-Inuletum verbascifoliae*. Other two were described for the first time. Data provided with this revision may be considered as an essential base-line information that should aid in evaluating the state of this vegetation type in the future.

**KEYWORDS:** *Asplenietea trichomanis*, *Centaureo-Campanuletalia*, cliff vegetation, phytosociology, syntaxonomy, Balkan

Oral presentation 18 04 11

## XEROTHERMIC CLIFF VEGETATION OF THE CENTRAL MEDITERRANEAN BASIN

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The xerothermic cliff vegetation of the Mediterranean Basin was originally included in the order *Asplenietalia glandulosi* (*Asplenietea trichomanis* class). Subsequently, due to the high regional diversification characterizing this vegetation type, other orders and numerous alliances have been proposed. The floristic, synchorological and syntaxonomic relationships among these syntaxa were in most cases defined on the basis of expert judgement and this led to different interpretations. This situation is more accentuated in the central Mediterranean, that is at the crossroad between eastern and western syntaxonomic units. The aim of this work is to revise the xerothermic cliff vegetation, from France to Greece. The revision was based on a data set of nearly 1250 relevés. The data set was hierarchical clustered, involving several data transformations. The dendrogram was interpreted from a syntaxonomic point of view, using nomenclatural type relevés as a basis. Indicator Species Analysis was used to identify the diagnostic taxa of the main syntaxonomic units and non-metric multidimensional scaling ordination was used to visualize the floristic relationships among syntaxa. Results showed three main clusters of relevés representing the chasmophytic vegetation of southern France and western Italy (*Asplenietalia glandulosi*, with 5 alliances), Adriatic area (*Centaureo-Campanuletalia*, with 4 alliances) and southern Balkans (*Onosmetalia frutescentis*, provisionally with only 1 alliance). The *Asplenietalia glandulosi* turned out to be characterized by a north-south differentiation that is represented by two suborders: *Asplenienalia glandulosi* and *Tinguarrenalia siculae*. The new proposed syntaxonomic scheme provides an overview of the cliff vegetation of the central part of the Mediterranean Basin and defines the floristic and chorological relationships among high-rank syntaxa, with new revised sets of diagnostic taxa. This revision might be useful for further detailed phytosociological works.

**KEYWORDS:** *Asplenietalia glandulosi*, *Centaureo-Campanuletalia*, classification of vegetation, Indicator Species Analysis, mediterranean cliff vegetation, *Onosmetalia frutescentis*



Oral presentation 19 04 54

### CLASSIFICATION OF *CARPINUS ORIENTALIS* DOMINATED COMMUNITIES IN BALKAN PENINSULA

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*Carpinus orientalis* is important tree species for many oak forest communities in Balkan Peninsula. However, many of these forests are degraded to *Carpinus orientalis* dominated scrub. These degraded communities where oaks are no longer important part of the upper layer, due to being mainly copices with similar bushy appearance, remained poorly known in terms of their floristic variability and ecology. This study aimed at, based on 728 relevés collected from all around Balkans using the standard Central European approach, resolving the syntaxonomy, ecology, nomenclature and distribution of these communities in Balkan Peninsula. Data were analyzed using numerical methods of classification and ordination in TURBOVEG, Juice, PC-Ord and R. Results show clear distinction of 16 communities grouped in Western-, Southern- and Central-Balkan clusters. Eight communities are already described associations, while another eight are potentially new associations. Following the traditional syntaxonomy, communities belong to thermophilous alliances of *Carpinion orientalis*, *Syringo-Carpinion orientalis*, *Fraxino orni-Ostryion*, and *Buxo-Syringion*. However, as some of the communities represent degradation of more mesophilous *Erythronio-Carpinion* and *Ostryo carpiniifoliae-Tilion platyphylli* forests with similar floristic composition as latter, their possible syntaxonomical position could be in some of these mesophilous alliances. We also question the grounds for the existence of the Central and Southern Balkan alliance of *Syringo-Carpinion orientalis*. This alliance brings together the oriental-hornbeam communities, which are all secondary succession stages of various types of oak forests originating from different alliances (*Carpinion orientalis*, *Quercion confertae*), which is not congruent with the syntaxonomical scheme of similar communities from Western Balkans where secondary succession stages of oak forests of *Carpinion orientalis* also belong to *Carpinion orientalis*. Our analysis has also shown that the upper layer of the so-called *shibljak* formations, albeit 'shrubby' in the appearance, is comprised of tree species with high cover value, so we argue

if those communities belong to scrub and mantle vegetation of *Buxo-Syringion* or to the forest vegetation of *Carpinion orientalis*.

**KEYWORDS:** Carpinetum orientalis, forest, phytosociology, scrub, syntaxonomy, vegetation

Oral presentation 20 04 52

### CHANGES BETWEEN CURRENT AND PAST COMPOSITION OF THE MACRO- AND MICROFLORA IN THE RICE FIELDS OF THE NATIONAL PARK OF THE DELTA OF AXIOS – LOUDIAS – ALIAKMON (N. GREECE)

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Rice fields, although artificial habitats, have great biological interest, because they function as substitutes of wetlands, supporting aquatic wildlife. In the National Park of the Delta of Axios – Loudias – Aliakmon they are widespread, covering c. 106.000 acres. In a previous study of macroflora in the same area, conducted in 1965, 36 higher plant taxa and one charophyte were recorded. A study of microflora carried out in 1976 reported 98 taxa belonging to three divisions, Bacillariophyta, Chlorophyta and Cyanobacteria. This work aims to reveal the differences between current and past composition of the macro- and microflora. We studied the macroflora in 2012 and 2016, based on a quadrat-sampling method and the microflora in 2016 from water samples taken from the same quadrats. In a total of 259 plots (4 m<sup>2</sup> each) examined, which were scattered all over the national park, 42 macroflora and 41 microflora taxa were recorded. The comparison to the previous study suggests that several macroflora taxa have disappeared (only 12 taxa are common in the two studies). They are indigenous aquatic taxa, e.g. *Schoenoplectismucronatus*, *S. litoralis*, *Typha latifolia*, *Juncus articulatus*. In contrast, a remarkable increase of alien taxa is observed. Among the 17 taxa we found, only four were recorded in 1965. The new alien taxa are mostly tropical, e.g. *Ammaniaauriculata*, *A. baccifera*, *Rotalaramosior*, *Heterantheralimoso*, *H. reniformis*. On the other hand, much less taxa of microflora were recorded compared to the study of 1976, probably due to the limited study period. However, our study revealed the occurrence of 28 taxa not previously recorded, several of which were classified to Euglenophyta, Cryptophyta, and Ochrophyta, also not known in those rice fields. The most frequent microflora taxa where the diatoms *Synedra ulna*, *Achnanthesbrevipes* var. *in-*

*termedia*, *Nitzschia* sp., *Navicula* sp., the chlorophytes *Chlamydomonas* sp., *Selenastrum* sp., the cryptophyte *Rhodomonas* sp., the cyanobacteria *Aphanothece* sp. and *Limnothrix* sp., and the euglenophytes *Euglena viridis* and *Phacus* sp. Our results show that both macro- and microflora have undergone great changes from the past, possibly due to the intensification of cultivation and new farming practices, e.g. the use of new herbicides, the introduction of new rice varieties etc.

**KEYWORDS:** National Park, rice fields, changes of the flora, weeds, cyanobacteria, microalgae

Oral presentation 21 04 20

### THE FLORISTIC COMPOSITION OF MUD VEGETATION IN DRIED WATER RESERVOIRS IN ALBANIA

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Over 700 0.1–10 ha large water reservoirs are known in Albania. Aside from scattered floristic records, their flora has not been systematically studied. The persistent drought of 2016–2017 in Albania offered an opportunity to examine the mud vegetation on the dried bottom of different-sized water reservoirs. Our objectives were to come up with a representative research regarding the flora of Albanian water reservoirs, to prepare a species list, to define the ratio of alien species and to examine the effect of the bedrock on the composition of the flora of these anthropogenic sites. Another question was how the flora of water reservoirs differs from the flora of natural water bodies and mud vegetation. We provided a list for each reservoir which contains the found species with their ratio. We have found 324 vascular plant species (almost 10% of the flora of Albania), 289 native and 35 adventive (17, 8% of the adventive flora of Albania); 161 annual/biennial and 163 perennial in 129 reservoirs. 4 of the 324 were new to the flora of Albania, these are: *Lindernia procumbens*, *Verbena supina* and 2 aliens: *Ammannia coccinea* and *Chenopodium pumilio*. There were 25,9 species per reservoir on average, the number of species varied from 0 to 76 in the individual reservoirs, and there were 124 species that we only found in one reservoir. In 119 reservoirs the coverage of vegetation was less than 30%. We have found that the flora of water reservoirs is regionally determined. These water reservoirs can be a refuge for plants that are connected to natural wetlands, as proof we found 158 such species. Thanks to migratory birds, these reservoirs can serve as a way for some species to spread and can provide habitat for rare plants, such as *Oldenlandia capensis*. On the other hand, grazing and manuring in and around reservoirs can be

a threat, as it can open the way for invasive species and nitrophilous plants. It has not escaped our notice that the ratio of Mediterranean and adventive species is worrying.

**KEYWORDS:** Albania, water reservoir, mud vegetation

Oral presentation 22 04 09

### MACROPHYTE HABITAT FRAGMENTATION IN THE MIDDLE COURSE OF THE DANUBE RIVER

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River networks are naturally fragmented by agents such as waterfalls, cascades, and beaver dams, but human disturbances have further divided these habitats. Anthropogenic disturbances operate on both a local and landscape scale to change river habitats. When acting jointly, localized and landscape-scale disturbances can have multiple, cumulative effects on aquatic habitat. Dams are an example of such a disturbance, and are known to have both localized and landscape-scale influences. We hypothesize that the physical features of river habitats and anthropogenic hydromorphological alterations influence macrophyte communities and lead to habitat fragmentation. Sampling included 1081 contiguous survey units positioned in the main channel and side arms along 588 km of the Danube River, along its middle course. To identify habitat fragments, Multivariate Regression Tree analysis (MRT) was applied on macrophyte and environmental data. Indicator species analyses were combined with MRT. To identify habitat fragments on a scale larger than final MRT groups, we set thresholds for an MRT complexity parameter. We identified 20 fine, 7 medium, and 3 large scale habitat fragments. Damming was found to be the main fragmentation agent. Macrophyte communities show continuous variation at all scales of habitat fragmentation. High species diversity indicates major anthropogenic alteration of the river's hydrology and decline of the natural riparian zone.

**KEYWORDS:** damming, hydrophytes, diversity, Multivariate Regression Tree

Oral presentation 23 04 31

### THE ROLE OF HYDROLOGICAL REGIME IN STRUCTURING MACROPHYTE ASSEMBLAGES IN GRAVEL PIT LAKES ALONG THE DRINA RIVER FLOODPLAIN (SERBIA)

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Hydrological regime plays an important role in structuring macrophyte vegetation in highly connected floodplain lakes, such as gravel pit lakes connected to the main river channel. If the dominant hydrological drivers are known, an optimal selection of gravel excavation location, relative to the main river channel, may create favorable hydrological conditions for aquatic biota. The aim of this study was to determine the most significant and relevant hydrological variables for structuring macrophyte assemblages in newly formed gravel pit lakes along the lower course of the Drina River. Field research was carried out on 49 survey sectors (14 gravel pit lakes), during the summer months of 2015 and 2016. Vegetation data was collected in accordance with the Pan-European standard for the sampling of macrophyte vegetation in lakes, using the UKTAG LEAFACS (Lake Assessment Methods, Macrophyte and Phytobenthos). Hydrological variables (frequency, duration and amplitude of the flooding events during the 4-year period prior to the vegetation survey) for each lake were extracted from the Republic Hydrometeorological Services according to the relative shore height to the Badovinci water-level station on the Drina River. The role of hydrological variables in structuring macrophyte assemblages was tested using partial Canonical Correspondence Analysis, while Generalized Linear Model was performed to test the ability of selected hydrological attributes to predict the macrophyte quantitative metrics (Shannon diversity index, species richness, total macrophyte cover, number of macrophyte functional groups, number of charophyte taxa and relative charophyte cover). The analyses showed that species richness, Shannon diversity index, total macrophyte cover and number of macrophyte functional groups were negatively predicted by the frequency of flooding events during the summer seasons in the past 4-year period. However, these macrophyte indices, including the number of charophyte taxa and the relative charophyte cover, were positively correlated with the frequency of flooding events during the spring periods. These results suggest that the frequency of flooding periods during

vegetation season may affect macrophyte vegetation structure by direct biomass removal. However, the frequency of spring flooding events influences the water trophic conditions which may increase macrophyte diversity and richness.

**KEYWORDS:** hydrology, gravel pit, lakes, macrophytes, Charophytes, floodplain

Oral presentation 24 04 64

### A PREDICTION FRAMEWORK FOR AQUATIC VEGETATION USING ARTIFICIAL NEURAL NETWORKS

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The ability to predict aquatic vegetation patterns, such as species composition, species richness and diversity based on various environmental variables may be useful to environmental resource managers and stakeholders. The aim of this study was to develop and validate predictive models for the macrophyte vegetation patterns using Artificial Neural Networks (ANN). The dataset included 56 macrophyte survey sectors collected from 14 gravel pit lakes along the lower course of the Drina River, during the summer months of 2015 and 2016. Predictive models were designed using macrophyte variables (species composition, shannon diversity index, species richness, total macrophyte cover, number of macrophyte functional groups, number of charophyte taxa and relative charophyte cover) as outputs and physico-chemical and hydromorphological parameters as inputs. Dataset was later reduced using Principal Component Analysis and Canonical Correspondence Analysis to select only significant variables. Models were made using Python programming language and Keras open source library for Artificial Neural Network support. Over 50 different ANN models were tested. The final model was performed on standardized data using Soft-Max activation function for outputs and included four layers. The highest prediction rate was obtained for vegetation composition which was presented with data matrix consisting of 27 species and 56 samples. Derived model could be used as a starting point toward exploring environmental sorting mechanisms of macrophyte assemblages.

**KEYWORDS:** Artificial Neural Network, gravel pit, vegetation, macrophytes

Oral presentation 25 04 40

### POLLEN AND CHARCOAL AS KEYS FOR UNDERSTANDING VEGETATION DYNAMICS DURING THE PAST - CASE STUDY FROM CENTRAL CROATIA

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Sediment from the biggest Croatian mire (Blatuša) was taken in 2015. Physical and geochemical analysis of sediments, qualitative and quantitative analyses of pollen palynomorphs and charcoal particles were performed. The following local zones were identified: *Pinus - Fagus - Ulmus* (Zone 1, depth 210-175 cm), *Fagus - Corylus* (Zone 2a, 175- 150 cm), *Fagus - Alnus* (Zone 2b, 150-85 cm), NAP (non-arboreal pollen) - *Fagus - Quercus* (Zone 3a, 85-45 cm) and NAP - *Carpinus - Quercus - Fagus* (Zone 3b, 45-5 cm). Local palynological species form following subzones: Cyperaceae - Polypodiales (210-180 cm), Polypodiales - *Sphagnum* (180-90 cm), *Sphagnum* - Polypodiales (90-40 cm) and Cyperaceae (40-5 cm). Zone 1 is characterised by 41 pollen types, the share of arboreal pollen is ~ 85%, the broader area was covered by pine and plants typical for oak forests, and today's area of the mire was a mosaic of wetland vegetation and wet meadows, with partially developed peatland vegetation. Zone 2 is characterised by 45 pollen types, the share of arboreal pollen is ~ 96%, with a domination of beech forest, the high share of hazel (Zone 2a) and alder (Zone 2b), and a local domination of ferns. Zone 3 is characterised by 57 pollen types, the share of arboreal pollen is ~ 72%, with a relative domination of grasses. Beech is the most common tree in Zone 3a and hornbeam and oak in Zone 3b, which is also characterised by the highest palynological richness and numerous anthropogenic indicators (eg. Cerealia pollen). According to the results of <sup>14</sup>C AMS dating, the deepest section of the core belongs to the Preboreal interval (Holocene). The shares of arboreal pollen, anthropogenic indicators, charcoal particles and palynological richness point to variable intensity of anthropogenic pressure, particularly expressed since the developed Middle Ages. Charcoal particles were the most numerous through the Zones 1 and 3, and are evidence of regional and local fires, essential succession factors. The results enable us to apply them for the same post-glacial period for the surrounding biogeographical area in the border zone between Central and Southeast Europe.

**KEYWORDS:** anthropogenic indicators, Blatuša peatland, fire, Holocene, paleoenvironment, palynology

Poster presentation 26 04 68

### NEW RECORDS OF ENDEMIC *IRIS ADRIATICA* (IRIDACEAE) IN NORTH ADRIATIC (CROATIA)

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*Iris adriatica* Trinajstić ex Mitić is the strictly endemic rhizomatous dwarf plant from the *I. pumila* complex, first recorded and described in 2002 in Croatia. Due to its small, lately decreased or disappearing populations, *I. adriatica* belongs to the NT (near threatened) IUCN category in the Croatian Red Book of Vascular Plants. Recent metabolic profiling of this species revealed its great pharmacological potential and chemotaxonomic relevance. So far, its recorded presence has been limited to the few locations in central Dalmatia (Croatia). Here we present the new records of *I. adriatica* on the Island Cres in North Adriatic. Two new locations were identified: one in the area of Srem, on the north of the church St. Mihovil (155 MASL) and the other, small population in the area of Jelovica, around the church of St. Juraj (141 MASL), which is the northernmost, so far confirmed record of this species in the Republic of Croatia.

**KEYWORDS:** *Iris adriatica*, new records, endemic species, Cres, North Adriatic

Poster presentation 27 04 18

### NEW SPECIES IN THE FLORA OF KOZARA NATIONAL PARK (BOSNIA AND HERZEGOVINA)

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According to the group of authors Bucalo et al. (2007) the flora of Kozara National Park comprises 737 plant species. During our floristic research conducted in the period 2012 – 2017., 51 new plant species in the flora of Kozara National Park were registered and the total species diversity of flora in this protected area now comprises 787 species. Out of 51 new species, 3 species are Pteridophyta, 36 species are Dicotyledonae and 11 species are Monocotyledoneae. There are 6 new allochthonous species (*Abutilon theophrasti*, *Euphorbia maculata*, *Reynoutria japonica*, *Lindernia dubia*, *Arum italicum*, *Vallisneria spiralis*) out of which 2 are invasive (*Vallisneria*

*spiralis*, *Reynoutria japonica*). There is one Balkan endemic species among new ones. According to structure two species are woody, three species are shrubs and others are *herbaceous*. Thirteen species inhabit aquatic, semi-aquatic and humid habitats out of which two species are aquatic macrophytes, whereas others inhabit an amphibian and riparian zone of rivers, streams and swamps.

**KEYWORDS:** Kozara, new species, flora

Poster presentation 28 04 82

### GERANIUM VERSICOLOR L., A NEW SPECIES FOR THE VASCULAR FLORA OF MONTENEGRO

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During floristic research in June 2017, a new species for the vascular flora of Montenegro - *Geranium versicolor* L. was found. This plant grows on the edge of thermophilous woods on the Sreteška Gora, above Sjevernica River canyon (left tributary of the river Morača) and near to the riverbank. The species *Geranium versicolor* L. is native in the flora of the South-Western Balkan (Albania, Greece, Montenegro, Serbia), Sicilia and Malta, and is naturalized in NW Europe (Britain, France and Ireland). In addition, a short morphological description of the *G. versicolor*, its population size on the mentioned localities, specific characteristics of the habitat, general distribution of the species and threatened status are presented.

**KEYWORDS:** vascular flora, Montenegro

Poster presentation 29 04 56

### THREE NEW ORCHID TAXA OF HYBRID ORIGIN FOR THE AREA OF SERBIA AND MONTENEGRO

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Among European food-deceptive orchids, hybridization between species from the same or different genera is a common phenomenon. Genus *Anacamptis* with c. twenty species is widespread in Europe, West Asia and North Africa. Many examples of natural hybridization among species from this

genus have been registered so far. During floristic investigations of Serbia and Montenegro from 2011 till 2018, three new hybrid orchid taxa (*A. × parvifolia*, *A. × timbali* and *A. × alata*) were registered. In all three cases, one of the parental species is from *laxiflora-palustris* group, while the other one is from *coriophora* or *morio* group. Hybridization of these species is a result of their similar geographical distribution, habitat requirements and flowering period. *A. × parvifolia* was registered for the first time in Montenegro in 2011, on Ulcinj Long Beach. Specimens of this hybrid and those of the parental taxa *A. coriophora* subsp. *fragrans* (about one hundred), lived on the fixed coastal dunes with herbaceous vegetation (*Onobrychis caput-galli* community). Nearby, second parent was found - *A. laxiflora*, also with a population of about one hundred specimens, but in different habitat - Mediterranean salt meadows (ass. *Juncetum maritime-acuti*). *A. × alata* was registered in 2014 in the vicinity of village Čanj (Montenegro). Specimens of all three taxa, both parents (*A. morio* and *A. laxiflora*) as well as hybrid, grew on stream bank, on artificially raised terrace overgrown in maquis vegetation. Only one hybrid specimen was found, while both parents were represented with several dozen specimens. The third hybrid taxon *A. × timbali* was registered in Special Nature Reserve "Selevenjske pustare" (Serbia) in May 2018. Eight specimens of this hybrid lived among few hundred specimens of both parents - *A. coriophora* subsp. *coriophora* and *A. palustris* subsp. *palustris*, in meadow vegetation (ass. *Rhinantho borbasii-Festucetum pratensis*). Appearance of hybrids among food-deceptive orchids is not a threat for the existence of parental species in research areas, as it was thought before. On the contrary, sympatric zones such as these could be important for the evolutionary processes in orchids, meaning that they could represent potential stimulus for evolutionary changes through promotion of local genetic adaptations.

**KEYWORDS:** *A. × parvifolia*, *A. × timbali*, *A. × alata*, food-deceptive orchids, sympatric zones

Poster presentation 30 04 51

### CROCUS HEUFELIANUS HERB. AND GONIOLIMON TATARICUM (L.) BOISS., TWO NEW SPECIES FOR THE FLORA OF ALBANIA, DISTRIBUTION AND ECOLOGY

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In Albanian Flora there are 19 taxa from the genus *Crocus* and *Crocus heufelianus* Herb. represents a new taxon for it, which occurs in northern and eastern Albanian Alps. It forms large population with more than thousand mature individuals in calcareous and shistous, subalpine and alpine pastures and meadows, in clearings between beech forests or in plac-

Poster presentation 32 04 27

### EXPANSION AND DIVERSITY OF HACQUETIA EPIPACTIS IN EUROPE

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As far as phylogeography of alpine and high mountain plants were intensively studied during last decades, plant geography of a mid or altitude woodland and herbaceous forest plant species was neglected. For this reason we paid more attention on *Hacquetia epipactis*, a herbaceous component of deciduous forest phytocoenosis. We studied the genetic structure of *H. epipactis*, using chloroplast regions and AFLP (fingerprinting method) as markers. We wanted to understand relations among widespread European populations (Dinaric Alps, Alps, Carpathians and Polish Lowland), and the main idea was to explore the phylogeographical relations in between *H. epipactis* populations and find the refuge areas for the northwards and eastwards migration of *H. epipactis*. We wanted also to answer on question, if its migration routes can be connected with postglacial migration of forest species, especially beech communities? In results, we found, that cp DNA showed a lack of genetic structure differentiation, that can indicate a relatively fast colonisation of *Haquetia* from the probably Balkan refugia. Another explanation is, that *H. epipactis* is a genetically and evolutionary stable genus that developed only one species. Results of AFLP fingerprinting suggest rather geographical isolation of studied populations, mainly by habitat fragmentation, thereby on limited gene flow. Additionally, we think, that probable is existing in the past, glacial refugia in Dinaric Alps or Balkan Peninsula and existence of the Holocene local hot spot in the Western Carpathians in Moravia. This localities in Moravia, was not a truly refugia, but rather a source of migration and postglacial colonisation to the north and eastern Europe (Poland). Closely situated populations that establish from a refuge area were more genetically similar. Our studies contradict, that an important role in expansion of Illyricoid species, such *H. epipactis* played *Fagus sylvatica*, as a dominant species of various communities with *H. epipactis*.

**KEYWORDS:** *Haquetia epipactis*, Balkan, AFLP, disjunction, migration

es forests of *Betula pendula* Roth, such as in Shishtaveci area. While *Goniolimon tartaricum* (L.) Boiss represents the second taxon besides *G. dalmaticum* Rchb. f. which was the only one taxon from the genus *Goniolimon* present in Albanian Flora books until now. The species was observed and collected in calcareous subalpine and alpine grasslands of eastern Albania. The identification of these taxa is based on a critical literature review and on comparison with specimens found at National Herbarium. Data on distribution, habitat and ecology are given for both species. Their presence increases the number of taxa in Albania and wide their distribution localities as well.

**KEYWORDS:** Albania, flora, new species, *Crocus heufelianus*, *Goniolimon tataricum*

Poster presentation 31 04 83

### NEW FLORISTIC RECORDS FOR THE FLORA OF THE REPUBLIC OF MACEDONIA

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The flora of the Republic of Macedonia is characterized by the diversity and richness of plant taxa. Because of this, despite the large number of surveys that have been carried out, new data on the distribution of some rare taxa in different regions on the territory of the Republic of Macedonia are still discovered. Investigation of the flora in the surroundings of the village of Krivogashtani (Prilep region) resulted with new data for the territory of the Republic of Macedonia. This contribution is a result of the processing of the herbarium material that was collected from the village of Krivogashtani and its surroundings, a lowland area located in the southwestern part of the country. The collection of the herbarium material was carried out during 2016/2017 within graduate thesis, according to the standard methods. In total, 52 families, 163 genera, 232 species were registered. Besides that, the area of distribution of 5 interesting and rare plant species was extended: *Bellardia trixago* (L.) All., *Stachys angustifolia* M. Bieb., *Tolpis umbellata* Bertol., *Romulea bulbocodium* (L.) Sebast. & Mauriand and *Carex disticha* Huds. Registration of the species *Polygonum arenarium* Waldst. & Kit, found in the eastern part of the researched area, along the Blato River, confirmed the data of Velenovsky (1922) about the existence of this species on the territory of the Republic of Macedonia. These data will contribute to the knowledge of the distribution area of these species from this part of the Balkan Peninsula.

**KEYWORDS:** new data, rare plant species, distribution area, Republic of Macedonia, Balkan Peninsula

## ENDEMISM IN THE UPLAND FLORA OF THE INVESTIGATED AREAS OF NORTHWESTERN SERBIA AND ŠUMADIJA

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The investigated areas in northwestern Serbia and Šumadija are part of peri-Pannonian Serbia, and cover 3,020 km<sup>2</sup> of land. The most important mountain massifs in this region are Povlen, Maljen, Suvobor, Ovčar, Kablar, Jelica, Vujan and Rudnik. The summits of these mountains reach altitudes ranging from 864 m (Suvobor) to 1,347 m (Povlen). Elevations range from 100 to 1,347 m. The major river in this part of Serbia is the Zapadna Morava (Western Morava) with its tributaries. The potential vegetation of this region forms two forest zones i.e. the continental deciduous thermo-mesophilous forest zone – *Quercion frainetto* zone, and the mesophilous forest zone – the *Fagenion* zone. The objective of the study was to make a list of Balkan endemic species and subspecies, present the chorological spectrum of the endemic flora of northwestern Serbia and Šumadija, and make a comparison with the endemic flora of Serbia and the Balkan Peninsula. Field surveys of the vascular flora of the region were carried out over the course of five consecutive years, and covered all stages of the growing season throughout the year. The results show the presence of 45 endemic taxa including species and subspecies, accounting for 3.37% of the total number of taxa in the region, which is comparatively much less than in the Balkans. The transregional Balkan endemites of the investigated region include 32 taxa (almost 70% of the total), 18 of which belong to the Eurasian Mountain Range (EMR). The regional endemites of the region include 9 taxa (about 20% of the total), 5 of which belong to the EMR. The local endemites of the region include 3 taxa (about 7.5% of the total), with *Knautia pancicii* belonging to the EMR, *Edraianthus canescens* to the Mediterranean/Submediterranean range, and *Salvia pratensis* subsp. *pozegensis* to the Pontic range.

**KEYWORDS:** endemism, northwestern Serbia, Šumadija, Balkan endemites, local endemites

## WHERE THE ENDEMICS ARE FOUND? DISTRIBUTION PATTERNS OF AREA-RESTRICTED VASCULAR PLANTS IN THE KARPATOS ISLAND GROUP (AEGEAN SEA, GREECE)

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Karpathos island group (Dodecanisos, Greece) at the south-eastern part of the Aegean Sea consists of 4 main islands (Karpathos, Kasos, Saria, Armathia) and many islets. Karpathos, the largest island, has a maximum length of approximately 49 km and covers an area of 302.2 km<sup>2</sup>. Its highest peak on Mt. Kalilimni reaches 1215 m. Geologically, the islands consist predominately of limestone, flysch and marls. From a phytogeographical point of view, the island group is important because it traditionally forms the border between the floristic regions of Europe and Asia. As currently understood, the flora of the area consists of at least 1100 taxa (species and subspecies), including several local and Aegean endemics. Distribution data has been collected from literature sources, various herbaria and our own geo-referred field work (2014-2016, 2018). A preliminary distribution matrix of 139 species and subspecies of local endemics, Aegean endemics or range-restricted taxa has been prepared. Three local taxa are new for the area (*Nepeta scordotis*, *Medicago arborea* subsp. *strasseri* and a hitherto undescribed Apiaceae member). A second supplementary data matrix includes potentially threatened taxa, as defined by the application of the IUCN criteria at national level. Locality data (altitude, substrate, habitat type, bioclimatic variables, etc) has also been added to the matrices. The Karpathos island group has been divided into 166 grid cells of 2 x 2 km and distributions within this grid map have been recorded. Smaller cell size is currently not recommended as it would be difficult to assess early floristic records, particularly those between 1960 to 1990. The aims of the project are to define: a) where the hot spots are located within the Karpathos island group and how are they related to conservation, b) how endemism is correlated with the selected environmental predictors and c) the phytogeographical relationships of the island group in the southern Aegean area.

**ACKNOWLEDGEMENTS:** the first author thanks SARG, National and Kapodistrian University of Athens, for funding his attendance to the Congress.

**KEYWORDS:** Aegean islands, chorology, conservation, phytogeography, plant diversity

## SOME ENDEMIC PLANTS SPREADING AROUND KARAGÖL AND ENVIRONMENT, IN BOLKAR MOUNTAINS (NIGDE-TURKEY)

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In this study, it was aimed to present some endemic plants distributed in the vicinity of 1-2 km of Karagöl Basin in Bolkar Mountains spreading within the boundaries of Niğde Province. In this research, approximately 37 endemic taxa belonging to 14 different families were collected in 2016-2017. Distribution data of these taxa and the ecological characteristics of the investigated area were determined and the digital photographs of the plants were taken. Two plant taxa were named after Bolkar Mountains (such as *Veronica bombycina* Boiss. & Kotschy subsp. *bolcardaghensis* M.A. Fisch., *Turanecio farfarifolius* (Boiss. & Kotschy) C. Jeffrey. Some of these plants species which are distributed around Karagöl in the Bolkar Mountains are; *Dianthus lactiflorus* Fenzl, *Salvia quezelii* Hedge & Afzal-Rafii, *Pedicularis cadmea* Boiss., *Lactuca glareosa* Boiss., *Linum empetrifolium* (Boiss.) P. H. Davis, *Potentilla pulvinaris* Fenzl. subsp. *argentea* Hartvig & Strid., *Verbascum tauri* Boiss. & Kotschy, *Cerastium gnaphalodes* Fenzl, *Pseudomuscari azureum* (Fenzl) Garbari & Greuter, *Silene fenzi* Boiss. & Balansa. In this study, biological data for approximately 37 plant taxa, of which some of them are locally endemic, were accomplished by field studies, herbarium samples, current literature and digital photos of the species.

**KEYWORDS:** ecology, endemic, local, Karagöl, Niğde, taxonomy

## THE ENDEMIC PLANTS IN BARTIN AND THEIR CONSERVATION STATUS

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Bartın, previously a district of Zonguldak located in the Western Black Sea Region of Turkey, earned a provincial status in 1991. Flora and vegetation studies in Bartın province has been started in detail since the second half of the 1990's. The first studies in this regard were carried out by Yatkin and Basa-

ran. After some small-scale studies carried out by some researchers in restricted areas in Bartın, a flora list for the whole province was published by Kaya and Basaran. According to Kaya and Basaran, there are 672 different plant taxa belonging to 368 genera under 97 families in Bartın. The authors stated that 7 of them (1.04 %) were endemic for Turkey. Afterwards, within the scope of Bartın's biodiversity project, Kaya and Yaman have worked on the flora of the whole province in detail. According to Kaya and Yaman, 33 of Turkey's endemic taxa are also found in Bartın's flora, and 7 of them are geophyte taxa. Recently, Bartın section of Kure Mountain National Park was also examined by other taxonomists in terms of its flora and vegetation. The conservation status of plants in the world has been determined by different organizations such as IUCN, CITES and BERN. The present study aims to evaluate the conservation status of endemic taxa in Bartın on the basis of IUCN, CITES and BERN criteria.

**KEYWORDS:** endemic, flora, vegetation, conservation status

## GENUS ALLIUM (AMARYLLIDACEAE, ALLIOIDEAE) IN SLOVENIA

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*Allium* is one of the most species rich (over 800 species), widespread and ecologically diverse plant genera, mostly confined to N Hemisphere. Representatives are easily recognized by characteristic smell (due to cysteine derivatives) and habitus. There are 21 known autochthonous species in Slovenia. Species richness increases towards the Submediterranean region. Gaps in our understanding of distribution of *Allium* species in Slovenia are mainly due to biased mapping. Rare and taxonomically critical species are searched for specifically, but are less likely traced or recognized in non-targeted field trips. A good part of the species is rare or distributed locally, possibly inhabiting extreme habitat types, sometimes with quite large populations. Plants at anthesis are not difficult to identify, however representation of taxa in herbarium collections and field lists is low due to short flowering period. Available determination keys exacerbate the problem by focusing on flowering time and thus make it difficult to accurately determine pre- (or post-) anthesis plants. In addition to some quite widespread and/or well known taxa, our revision has been focused in some taxonomically critical groups. *A. angulosum* and *A. lusitanicum* are morphologically quite similar, but easily discernable by their habitat preferences: inhabiting seasonally flooded meadows, and dry stony grasslands on limestone, respectively. *A. kermesinum* is a steno-endemic species of Kamnik-Savinja Alps. For *A. suaveolens* there are only two known populations, both on wetlands, one in brackish and the other in freshwater conditions. These populations may

possibly exhibit a degree of taxonomic variance. *A. roseum* and *A. neapolitanum* are calciphobous and thus limited to the flysch of the Adriatic Coast region, with a likelihood of *A. neapolitanum* population being a relict of former cultivation. *A. oleraceum* is widespread but rare, possibly overlooked, due to its similarity with *A. carinatum* and a tendency to reproduce only vegetatively in shaded areas. *A. scorodoprasum* subsp. *rotundum* and *A. sphaerocephalon* subsp. *sphaerocephalon* share somewhat similar habitus and habitat preferences, however are easily distinguished by their leaves: the first having flat and the other fistulose leaves. In *A. paniculatum* agg. at least two species exist in Slovenia: *A. dentiferum* had not been recorded earlier and *A. pallens* subsp. *pallens* which was recorded just recently after a long period.

**KEYWORDS:** *Allium*, flora of Slovenia

Poster presentation 38 04 32

### CONTRIBUTION TO THE KNOWLEDGE OF THE GREEK *ALLIUM*: WHAT *ALLIUM ACHAIUM* BOISS & ORPH. IS?

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According to the more recent literature related to the Greek Flora, *Allium* is represented in Greece by more than a hundred species with, almost 50% of them being Greek endemics. It is worth noting, however, than three only of these endemics viz. *Allium frigidum* Boiss. & Heldr., *A. achaium* Boiss. & Orph. and *A. parnassicum* (Boiss.) Halacsy, are exclusively adapted to higher altitude habitats (mountains of Sterea Ellas and Peloponnisos). Obviously, these three species show morphological similarities, since, in the past, by well experienced Botanists, *Allium parnassicum* was classified as a variety of *A. achaium* (*A. achaium* Boiss. & Orph. var. *parmassicum* Boiss.) and *A. achaium* was treated as a synonym of *A. frigidum*. The last four years, a thorough floristic exploration of Mt. Klokos in NW Peloponnisos (the *locus classicus* of *A. achaium*) showed that from the above three species, only *A. frigidum* occurs in the mountain concerned. Also, a detailed study of the original collection of Orphanidis from the same mountain, in which the original description of *A. achaium* is based, revealed that it consists of material belonging to *A. frigidum* too. As, however, the taxonomical approach recommended in the “*Mountain Flora of Greece*” i.e. the recognition of the above three taxa as distinct species, is accepted by the present authors, the necessary taxonomical and nomenclatural changes are recommended.

**KEYWORDS:** *Allium*, endems, Greek Flora

Poster presentation 39 04 73

### NOTES ON THE FLORA OF BELARUS: THE GENUS *ARABIDOPSIS*

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The genus *Arabidopsis* (DC.) Heynh. (*Brassicaceae*) is a small genus containing 9–15 species. This genus is of great interest since it contains *Arabidopsis thaliana*, one of the model organisms used in studies of plant biology. In Belarus this genus contains at least three native species. 1) *Arabidopsis thaliana* (L.) Heynh. It has widespread distribution in Belarus. 2) *Arabidopsis arenosa* (L.) Lawalrée s. l. The species is extremely polymorphic both in the whole distribution area and on the territory of Belarus, and requires detailed study at the morphological, karyological and molecular levels. Within this group some di- and tetraploid races are distinguished, most of which have a local distribution, mainly in the mountains of Central Europe. These races most often have geographic and/or reproductive isolation and should be viewed as species. In Belarus, this complex has not been studied. At the same time, some specimens, mainly from the northern and northwestern regions of the republic, have very weakly pronounced teeth at the base of the petals or none at all, approaching in appearance to the North American *A. lyrata*. Primarily in the western and central regions of Belarus there are perennial plants with a very dense and long pubescence of the stem and leaves, often having strongly dissected basal leaves with a small apical lobe. Such plants are externally similar (perhaps even identical) to the Pannonian *A. petrogena*. In the northwest of Belarus, almost glabrous perennial plants were collected, which are morphologically similar to the Carpathian *A. neglecta*. This group requires revision in Belarus. 3) *Arabidopsis suecica* (Fries) Norrl. This allotetraploid species originated from the hybridization between *A. thaliana* and *A. arenosa* s. l. Currently, *A. suecica* is known mainly in Fennoscandia. The modern methods of molecular research demonstrated that this species arose about 14 Kya, when the territory of Scandinavia was covered with an ice shield. In Belarus this species has not been reported before, although it was known from Poland, the Baltic states and North-West Russia. We discovered it in several localities in the north-western regions of Belarus. Probably, the species is much more widespread in these areas but has been overlooked.

**KEYWORDS:** *Arabidopsis*, Belarus, variation, distribution

Poster presentation 40 04 86

### DISTRIBUTION AND OCCURRENCE OF *ARBUTUS* L. TAXA (ERICACEAE) IN CROATIA

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The genus *Arbutus* L. includes circa 20 taxa that are distributed in the Mediterranean area, North and Central America. In wider Mediterranean region the genus is represented with four evergreen woody species: *A. unedo* L., *A. andrachne* L., *A. canariensis* Duhamel, *A. pavarii* Pump. and two natural hybrids (*A. × andrachnoides* Link and *A. × androsterilis* Salas, Acebes & del Arco). In Croatian flora according to Flora Croatica Database only three taxa occur along the coast and islands: *A. unedo*, *A. andrachne* and their hybrid *A. × andrachnoides*. All three taxa are typically Mediterranean element that have important role in formation of different types of macchias. The aim of this survey was to re-evaluate the occurrence and distribution of *Arbutus* taxa in Croatia based on literature data, field observations and herbarium specimens. Herbarium revision was performed and based on leaf, stem and fruit morphology on specimens deposited in CNHM, ZA, ZAGR and ZAHO. Results of this survey showed that *A. unedo* is very frequent, while *A. andrachne* and *A. × andrachnoides* are very rare with very restricted distribution and sporadic occurrence on few Adriatic islands. The widespread species is *A. unedo* which is distributed from Istrian peninsula on north to Prevlaka on south, and it can be seen that *A. unedo* is connected to very narrow area of the Croatian littoral with a very frequent occurrence on the islands. According to literature data, *A. andrachne* occurs on the islet of Badija, islands of Korčula, Kornati, Šolta, Mljet, and in the botanical garden of Lokrum. The occurrence of *A. andrachne* was not confirmed for the islet of Badija, neither for the islands of Korčula and Mljet, but we have confirmed the occurrence of hybrid taxon *A. × andrachnoides* with low number of individuals in population. It is questionable if *A. andrachne* still really grows on those islands. The occurrence of *A. × andrachnoides* is confirmed for the islands of Korčula, Mljet, Šolta and Lokrum, but not on the island of Vis. On the islands of Mljet and Korčula, where *A. unedo* and *A. × andrachnoides* grow together, individuals of probably introgressive origin were also found.

**KEYWORDS:** Adriatic, *Arbutus*, flora, herbarium, hybrids, morphology, systematics

Poster presentation 41 04 87

### ON THE DISTRIBUTION OF *CAMPANULA* SPECIES OF SERIES *GARGANICAE* TRINAJSTIĆ IN ALBANIA, ECOLOGY AND HABITAT PREFERENCE

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*Campanula* L. is one of the richest genus in Albania with ca. 40 species; out of them, six species, *C. aureliana*, *C. comosiformis*, *C. daucoides*, *C. korabensis*, *C. longipetiolata* and *C. skanderbegii* are known so far as Albanian endemics. The literature data show that the *Campanula* genus, series *Garganicae* Trinajstić in Albania is represented from two endemic species, *C. aureliana* and *C. skanderbegii*, known so far to occur only in their *locus classicus*. The research aimed: (i) to defined the distribution range of the species of series *Garganicae* in Albania; (ii) to identify the variability of morphological characters in different populations of the observed species and (iii) to determine the habitat type of the species occurrence, its ecological requirements as much as the population size and area of occupation. Twenty field observations were carried out during years 2016 and 2017, focused in the known localities and along the main deep canyons and gorges of Albania. Living plants were examined in the field and at list 10 plant species for each locality are collected, dried and herbarised. Dried specimens were compared with the specimens deposited in the National Herbarium of Tirana (TIR!) and Kew herbarium (K!). The identification of new species for the flora of Albania was based on both dry and living material. The field observations result in the identification of *Campanula debarensis* Rech.f as a new species for the flora of Albania. Further analysis showed also that this species, together with *C. aureliana* and *C. skanderbegii*, occurs in several others localities, unknown up to now. All Albanian species of series *Garganicae*, have wide distribution ranges, from 320 m up to 2000 m above sea level; most of them are found in calcareous rocky slopes with chasmophytic vegetation.

**KEYWORDS:** Albania, *Campanula* series *Garganicae*, *C. debarensis*, distribution range

**GALIUM L. IN NORTHERN PART OF SERBIA – DIVERSITY AND DISTRIBUTION**

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*Galium* L. is one of the largest genera of the tribe Rubieae Baill, family Rubiaceae Juss.. It comprises more than 600 species, mostly occurring in temperate areas, but also in alpine and arctic regions to subtropical and tropical zones at higher elevations. In the Flora of Europe, there are 145 *Galium* species registered, whereby 30 of them in the flora of Serbia. Given that Serbia has two clearly defined geographical units, the aim of this research paper was to determine the number of species of this genus and their distribution in a potentially poorer unit such as Northern i.e. Pannonian Serbia. Only verifiable herbarium data was used to form a database. Considering beforementioned, four herbarium collections with the largest number of specimens from Pannonian Serbia were examined: Herbarium of Faculty of Sciences, University of Novi Sad (BUNS), Institute for Nature Conservation of Vojvodina Province (PZZP), Herbarium of the Institute of Botany and Botanical Garden “Jevremovac”, University of Belgrade (BEOU) and the Herbarium of Natural History Museum in Belgrade (BEO). The total number of 844 herbarium sheets was examined, of which 484 are deposited in the collection of BUNS, 235 in PZZP, 64 in BEOU and 61 in BEO. Identification and revision of the herbarium material was made according to Flora Europaea and the regional Floras relevant for the investigated genus, while nomenclature alignment was done using the latest literary and online sources. The presence of 19 species and one subspecies from six sections of the genus *Galium* was determined: sect. *Leiogalium* (7 species and 1 subspecies), sect. *Kolgyda* (5), sect. *Aparinoides* (3), sect. *Platygalium* (2), sect. *Hylaea* and sect. *Galium* (1 species). The distribution of each *Galium* species/subspecies in Vojvodina was presented on the grid map with squares 10 × 10 km, based on Universal Transverse Mercator (UTM) projection. The least number of the herbarium data was recorded for species *G. divaricatum* and *G. tenissimum* from section *Kolgyda*. Material revision opened many questions considering diversity of this group in the area of Vojvodina, among others the status-obscure taxa *G. spurium* which was recorded throughout 13 exsiccates in examined herbarium collections.

**KEYWORDS:** *Galium*, diversity, distribution, revision, Pannonian Serbia, herbarium collections

**CONTRIBUTION TO THE FLORA OF THE LONG BEACH AND ITS HINTERLAND IN UL-CINJ (MONTENEGRO)**

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Ulcinj is located in the outermost south-eastern part of the Montenegrin coast and its openness towards the Adriatic and Mediterranean Sea (Otrant Gate) as well as the Mediterranean climate influence the diversity of flora and vegetation in this area. Based on the analysis of previous scarce botanical data of the Long Beach area and the island of Ada Bojana, we have performed a more detailed floristic research in this area. The explored terrain covers an area of 1,500 ha and includes 13 km long Long Beach and the island of Ada Bojana 3,7 km in length. Field investigations as well as the review of the literature concerning the Long Beach area and Ada Bojana island registered 962 taxa classified in 125 families and 516 genera. The family with the largest number of taxa is Poaceae and the genus with the largest number of taxa is Euphorbia. By analyzing the life forms, the dominance of therophytes and hemicryptophytes was revealed, explained by the geographical position of the investigated terrain, ie, the intense influence of the Mediterranean climate. Phytogeographic analysis showed that the most common type is the Mediterranean-submediterranean areal type due to the strong influence of the Mediterranean climate. On the Long Beach and the island of Ada Bojana there are 12 NATURA 2000 habitats and several associations that are classified in a particular type of habitat. The background of the beach is dominated by Mediterranean salt marshes (*Juncetalia maritimi*), Mediterranean temporary ponds, Mediterranean high hydrophilic meadows (*Molinio-Holoschoenion*) and white cannon and white poplar galleries, while the beach itself is dominated by the rudiments of mobile coastal dunes, mobile coastal dunes from *Ammophila arenaria* (white dunes), fortified coastal dunes with green vegetation (gray dunes) and inland pastures with one-year species (*Brachypodietalia*). 16 endemic, 36 relicts and 37 protected taxa were identified. The conclusion of this longterm research is that the anthropogenic factor has the most negative impact on the flora and vegetation of the investigated area, the most dangerous being urbanization, tourism, sand exploitation, waste disposal and introduction of allochthonous species.

**KEYWORDS:** Long Beach, flora, taxonomic, ecologic, phytogeographical study

**DISTRIBUTION AND DIVERSITY OF ORCHIDS IN WESTERN SERBIA**

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Understanding how patterns of orchid species richness and distribution vary along geographical and ecological gradients is fundamental for their conservation. Although the Balkan Peninsula is one of the parts of Europe with the highest number of orchid taxa, the orchid flora in the central Balkans is not sufficiently studied. The main objectives of this study were to determine the orchid species richness and diversity centres in western Serbia and the factors influencing the patterns of their distribution. Database was filled by chorological data from personal field investigation in the period 1995-2017, herbarium material from BEOU and BEO collections, and published data. The results of diversity analysis and summary distribution of orchid taxa were presented on 10×10 km and 50×50 km grids using Universal Transverse Mercator (UTM) projection. The presence and composition of orchid taxa in three geographical provinces and 11 mountain areas in western Serbia were also analyzed. Regression analysis was used to explore the relationship between orchid species richness and the environmental variables (altitude, 19 bioclimatic variables) and habitat heterogeneity within each UTM 10×10 km grid cell. The analyses were performed for all orchid taxa, and then for subsets, classified according to division of life forms based on orchid root characteristics. In total, the presence of 57 orchid species and subspecies in the investigated area was recorded. The most widespread species are *Anacamptis morio* (91 UTM 10×10 km grid cells), *Gymnadenia conopsea* (81 cells) and *Neottia nidus-avis* (69 cells). The results demonstrated that the largest number of orchid taxa are present in western Serbia, followed by the geographical provinces of southwestern and northwestern Serbia. Mt. Tara represents a distinctive centre of richness and diversity of the orchid flora in western Serbia. Moreover, the pattern of orchid richness was considerably shaped by the elevational gradient and habitat heterogeneity. Furthermore, the strongest influence of altitude (maximum values) and bioclimatic variables (minimum values of temperature variables) were determined for orchids with palmate and fusiform tubers. The results of this study may contribute to a better understanding of potential effects of habitat and climate changes on orchid diversity, and the conservation planning.

**KEYWORDS:** Orchidaceae, distribution, species richness, elevational gradient, biogeography, Balkan Peninsula

**CONTRIBUTION TO THE KNOWLEDGE OF ORCHID FLORA OF THE ȚARCU MOUNTAINS (SOUTHERN CARPATHIANS, ROMANIA)**

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The Țarcu Mountains form the western part of the larger Retezat-Godeanu-Țarcu massif, which is itself the westernmost part of the Southern Carpathians of Romania. They reach their highest points in the Vârful Pietrii (2192 m) and Țarcu (2190 m) peaks, thus being lower than the neighboring Retezat Mountains. The field researches regarding the orchid's family in the Țarcu Mountains area (the species inventory, the inventory, distribution, size and dynamics of populations as well as the acknowledgment of the threats with impact upon the orchid species and populations and on their habitats) have been began in the summer of 2013. Thanks to easy access, although it is classified as part of the European Natura 2000 network (ROSCI0126 Munții Țarcu), was supported strongly negative anthropogenic impact factors. Although considered in most part anthropized, field research, it was concluded that there is growing 12 species of orchids.

**KEYWORDS:** orchids, conservation, threats, Țarcu Mountains, Romania, Orchidaceae

**FLORISTIC DIVERSITY OF MOUNTAINS OLIGIRTOS AND FARMAKAS (NORTH-EAST PELOPONNISOS, GREECE): PRELIMINARY RESULTS**

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Peloponissos, a large peninsula at the southernmost part of the Greek mainland, is one of the richest floristic regions of Greece in endemic taxa: approximately 22% of its species and subspecies are endemic. Several mountains in this area have been thoroughly investigated, among them Killini, Erimanthos and Panachaiko. Still, floristic gaps do exist, especially along a diagonal line from the north-east to the south-west of the peninsula. Two neighbouring mountains, Oligirtos (1935 m) and Farmakas (1615 m), have been selected for study because their flora is poorly known. In the literature, there are 87 confirmed taxa records from Oligirtos and only 6 from Farmakas. Furthermore, the largest part of Mt. Oligirtos is included in the Natura 2000 network and a good study of its

flora is crucial for conservation. The geology of the mountains comprises mainly limestone, dolomite and flysch. Since March 2016, 12 daily field trips have been carried out to Oligirtos and 13 to Farmakas. A total of 1634 specimens have been collected from the mountains: 694 from Oligirtos and 940 from Farmakas. Up to date, 501 taxa have been identified from the study area; 334 taxa from Oligirtos, belonging to 199 genera and 66 families and 321 taxa from Farmakas, belonging to 195 genera and 65 families. Only 154 taxa are found on both mountains. The local endemic *Stachys chrysantha* forms its northernmost populations on Oligirtos and presents unusual white flowers. *Tripodion graecum* is distributed on both mountains and shows a disjunct area of occurrence, shared between a few mountains of Peloponnisos, south Turkey and northern Syria. This is quite similar to the distribution range of *Biebersteinia orphanidis*, a rare plant recorded on Oligirtos. Fourteen local endemics have been found on the mountains so far, 5 on Farmakas and 12 on Oligirtos. Among them, *Asperula arcadiensis* (both mountains), *A. saxicola* (Farmakas), *Adonis cylleneae* and *Viola oligyrtia* (Oligirtos). The study is being carried out in the framework of a PhD degree and will continue the following years.

**ACKNOWLEDGEMENTS:** the first author thanks SARG, National and Kapodistrian University of Athens, for funding his attendance to the Congress.

**KEYWORDS:** endemism, mountain flora, plant distributions, NATURA 2000

Poster presentation 47 04 39

### FILLING THE FLORISTIC GAPS IN THE AEGEAN: THE CASE OF DOKOS ISLAND (MIRTOŌN ARCHIPELAGO, GREECE)

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Mirtoön Sea and its archipelago define the south-western parts of the Aegean Sea and consist of 10 islands of various sizes and several islets or island rocks. In decreasing order of size Idrá, Spetses and Dokos are the largest ones, while Falakona and Velopoula are those most remote from the mainland. Only the two largest ones are permanently inhabited. Dokos covers an area of 13.5 km<sup>2</sup> and lies roughly between Idrá and Spetses. It is separated from Peloponnisos by a strait of c. 0.95 km. Its northern coast is mild, forming the large gulf of Kouata but its southern coastal part is precipitous and inaccessible. The island is predominately calcareous, with a maximum altitude of 308 m. Compared to the neighbouring Idrá (423 species and subspecies of vascular plants are known), Spetses (512 taxa) and Trikeri (90 taxa), Dokos is almost totally neglected: only 5 taxa have been recorded so far. To cover

this floristic gap, field work was conducted in 2018, resulting in 325 collected plant vouchers and several notes, representing at least 290 taxa of vascular plants. The vegetation of the island consists mainly of evergreen sclerophyllous shrubs of the thermo-Mediterranean zone dominated by *Juniperus phoenicea*. There are also phrygana formations, while in coastal and inland rocky areas there are halophytic and chasmophytic communities, respectively. Parts of the island in the north have been subject to cultivation and some rare weeds (*Bupleurum lancifolium*, *Convolvulus pentapetaloides*) still grow in open fields or olive groves. A few local taxa of the nearby East Peloponnisos also grow on Dokos, mostly on calcareous rocks (*Campanula andrewsii*, *Stachys swainsonii* subsp. *argolica*). Most of the floristic diversity recorded on Dokos is also present in Argolis peninsula (at least 1096 taxa provided in the literature), an expected assumption given the proximity of the two areas. Still, a few taxa found on Dokos (e.g., *Ajuga iva*, *Arenaria muralis*, *Catananche lutea*, *Diploaxis muralis*, *Hep-taptera colladonioides*, *Trifolium subterraneum*) have not been recorded in other islands of the Mirtoön archipelago.

**ACKNOWLEDGEMENTS:** the first author thanks SARG, National and Kapodistrian University of Athens, for funding his attendance to the Congress.

**KEYWORDS:** floristics, plant diversity, Aegean islands, Mirtoön Sea

Poster presentation 48 04 35

### DISTRIBUTION AND ECOLOGY OF MYCO-HETEROTROPHIC PLANTS IN THE ALBANIAN FLORA

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Mycro-heterotrophic plants are partly or entirely non-photosynthetic plants that obtain energy and nutrients from fungi. These plants form a symbiosis with arbuscular mycorrhizal, ectomycorrhizal or saprotrophic fungi to meet their nutrient demands. The current knowledge on the global extent of myco-heterotrophy includes 400 species of fully myco-heterotrophic plants and nearly 20 000 species of partially myco-heterotrophic. Research on this group of plants in Albania is almost totally missing. The aim of this study is to summarize the information existing about the myco-heterotrophic plant species found so far in Albania. The data for this research have been gathered through literature review and visiting the Albanian National Herbarium. Results show that, a total number of 14 species of myco-heterotrophic plants, from two families, specifically Orchidaceae and Ericaceae have been found up to now in Albania. The Orchidaceae family includes 8 species which have lost the ability of the photosynthesis process and

have totally reduced and lost their leaves. On the other hand the Ericaceae family is represented by 6 species, all belonging to Monotropoideae sub-family, and some are partially myco-heterotrophic and others are fully myco-heterotrophic species of tribe Pyroleae and Monotropeae respectively. Myco-heterotrophy is one of the longest-studied aspects of the mycorrhizal symbiosis, but there remain many critical, unanswered questions regarding the ecology and physiology of myco-heterotrophic plants and their associated fungi.

**KEYWORDS:** Albania, myco-heterotrophic plants, distribution

Poster presentation 49 04 42

### VASCULAR FLORA OF RICH FENS IN KOSTROMA OBLAST (EUROPEAN RUSSIA)

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Rich fens in European Russia represent ecosystems of high biological significance; though remain less investigated in comparison with central-European ones. Traits of vascular flora provide a basic knowledge for delimitation of different vegetation types of fens along the poor-rich gradient within a certain area. The study aims to survey vascular flora of rich fens of Kostroma Oblast, the least studied region of central part of European Russia, with focus on species-rich habitats and sites inhabited by rare plants. In this study, floristic and vegetation data were collected in 2015–2017 in three large undisturbed and slightly disturbed valley fen complexes in Kostroma Oblast (Upper Volga region). These fens characterised by specific water chemistry, mosaic structure of plant cover and rich vascular flora. pH of ground water tends to be from slightly acidic (less than 7.0) to slightly basic (about 8.0). Mire vegetation is similar to *Sphagnum warnstorffii*-*Tomentypnion nitensis* and *Caricion davallianae* alliances, which are represented in central-European rich fens, but has regional specific characters. These plant communities provide habitats for a number of rare species, like *Angelica palustris*, *Liparis loeselii*, *Pedicularis sceptrum-carolinum*, *Saxifraga hirculus*, etc. Vascular flora of rich fens investigated comprises 175 species with Cyperaceae and Orchidaceae as largest families. These ecosystems contribute markedly to the overall vascular plant species composition of mires of Kostroma Oblast. The species richness of different habitats is assessed, though correlation with pH was not revealed. It is supposed that species richness mostly depend on local mosaic structure of fen vegetation (e.g. presence or absence of strong dominant species, like *Scirpus tabernaemontani* and *Phragmites australis*). Most species-rich habitats are represented among sedges-herbs-brown mosses communities. Presumably vegetation of the fens belongs to three types along the poor-rich gradient: moderately rich fens, rich fens and extremely rich fens. Current data

support great biological and conservation value of these fens. They are able to maintain high species diversity and rare plant communities and might be considered as wetlands of interregional significance. Further research is needed to assess habitat diversity in these fens. Data obtained within the study are useful for comparison with data from European countries.

**KEYWORDS:** mineral-rich fens, mire, poor-rich gradient, rare plants: species richness

Poster presentation 50 04 61

### ECOLOGY AND SYNTAXONOMY OF THE HABITAT OF THE RARE SPECIES LATHYRUS TRANSILVANICUS IN BULGARIA

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*Lathyrus transsilvanicus* (Spreng.) Fritsch (Fabaceae) is a threatened species in the Bulgarian flora included in the Red Data Book of the Republic of Bulgaria, Vol. 1 under the category "Regionally Extinct". It was firstly found in 1903 in Central Balkan Range at 1600 m altitude and had not been confirmed until 2007, when it was found over again and its status was reassessed according to the criteria of IUCN Red List Categories and considered as "Critically Endangered". The species is distributed in Central Europe with southern limit in the Balkan Peninsula. In Bulgaria it has a very limited distribution in Triglav massif - Central Balkan Range, occurring in only 3 close to each other locations and presented by several individuals. Less is known about the ecological and floristic features of the communities of its habitat. The study aims to reveal the ecological peculiarities of the habitat of this rare species and to propose a phytosociological classification of its communities. The habitat characterization of *L. transsilvanicus* is made according to the principles of Braun-Blanquet phytosociological methodology. Several relevés were made in the established locations of the species and in some neighboring ecologically and floristically similar communities in order to obtain more complete syntaxonomical picture of its type of vegetation. In the Central Balkan Range *L. transsilvanicus* participates in tall-herb communities of *Molinia coerulea* and *Calamagrostis arundinacea* along streams at about 1550 m a.s.l. always near the tree-line of the beech forest belt of the region. These communities form a specific habitat for the species strictly following the forest canopy at the tree-line and the water along the streams. The slopes are steep with predominant southern exposure and limestone or very rarely silicate bedrock. Syntaxonomically these communities belong to the marginal vegetation of class *Mulgedio-Aconitetea* Hadač et Klika in Klika et Hadač 1944. Some comparisons with

the type of vegetation and the habitat of the species in other locations of its distribution range in Central Europe are also made.

**KEYWORDS:** *Lathyrus transilvanicus*, habitat, *Mulgedio-Aconitea*, syntaxonomy

Poster presentation 51 04 29

### MONITORING OF THE NATURAL HABITATS ALONG AN OXBOW OF THE DRAVA RIVER

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There is a widespread lack of assessment of riverine habitats previous to the restoration measures, as well as lack of monitoring after the project implementation. Our study is an effort to establish monitoring and collect baseline data. Habitats of the Old-Drava oxbow were surveyed in the period of 2015-2018. A habitat map was produced using the categories of the Hungarian national habitat classification system. Along the 17 km long oxbow 76 habitat patches were differentiated, representing 17 habitat types. The most significant Natura2000 habitats were: 3150 Natural eutrophic lakes with Magnopotamion or Hydrocharition type of vegetation; 91E0 Alluvial forests with *Alnus glutinosa* and *Fraxinus excelsior*. The objective of the investigations was to detect the magnitude and direction of the effects of water regime changes on Natura2000 habitat types with as high accuracy as possible. The method of permanent plots suggested by the Hungarian National Biodiversity Monitoring System was applied. For aquatic vegetation the coenological samplings were recorded in 4m<sup>2</sup> plots, arranged in five transect lines across two waterbed sections. Three different stands of gallery forests (white willow gallery; white poplar gallery; alder gallery) were assessed on 30×30m plots, while sampling of the herb layer was performed at a much higher resolution: in 55 quadrates of 0.5 m<sup>2</sup> area each, the percentage cover values were estimated for each species. Our results indicated that the aquatic vegetation is highly valuable and undisturbed: the proportions of the indicator species and the protected species signifying the naturalness of water plant associations were found to be high (27-33%, 17-30%, respectively) in all of the lines. Increasing water depth resulted in higher vegetation cover both in the submersed and floating plant strata, while species number and species density was not changed. In the forest stands we also detected favorable signs of naturalness: high species number (28-42 vascular plant species /900 m<sup>2</sup>), high proportion of species typical of the sampled forests (14-28%); and a small proportion of weeds (3.7-9.7%) and invasive plant species (6%). Our results also suggested a higher capability of the forest stands with higher species diversity to adapt to changes in climatic conditions.

**KEYWORDS:** coenological sample, naturalness, permanent plot, riverine habitat, water regime

Poster presentation 52 04 23

### THE MEADOW VEGETATION IN THE SOUTHWESTERN PART OF THE REPUBLIC OF MACEDONIA (THE REGION OF DEBRCA)

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The lowland meadow vegetation in the southern Balkan is classified within the class *Molinio-Arrhenatheretea* Tx. 1937, order *Trifolio-Hordeetalia* Horvatić 1963 and alliance *Trifolion resupinati* Micevski 1957. The aim of the study is to identify the characteristics of lowland meadow vegetation in southwestern part of the Republic of Macedonia - Debrca region (between the cities of Ohrid and Kichevo). We compared total 150 relevés from literature sources and 50 unpublished with own 38 relevés. Agglomerative hierarchical cluster analyses have been made in program PC-ORD. The influence of site and climate on species composition was shown in applied Pignatti indicator values and climatic data. Different life forms were analyzed statistically and through a detailed analysis of area-spectrum, we got a rather complete phytogeographic structure of the meadows at the researched area. From already described five lowland meadow plant associations in the Republic of Macedonia within the alliance *Trifolion resupinati*, in the research area were registered three: *Cynosuro-Caricetum hirtae* Micevski 1957, *Trifolietum nigrescentis-subterranei* Micevski 1957 and *Trifolietum resupinati-balansae* Micevski 1959. The analysis of life forms confirmed the therophyto-hemicryptophytic physiognomy of the lowland meadow associations within the alliance *Trifolion resupinati*.

**KEYWORDS:** *Trifolion resupinati*, meadow vegetation, Republic of Macedonia

Poster presentation 53 04 46

### MARSHLAND VEGETATION (PHRAGMITO-MAGNOCARICETEA KLIKA IN KLIKA ET NOVÁK 1941) OF THE CENTRAL BALKAN PENINSULA: FLORISTIC DIFFERENTIATION OF ASSOCIATIONS

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Vegetation class *Phragmito-Magnocaricetea* is considered to be one of insufficiently studied vegetation types in the area of Balkan Peninsula, in terms of composition, structure, distribution and ecology. In order to determine syntaxonomic relationships within the studied vegetation class, we conducted hierarchical classification analysis (UPGMA) to a data set that combines so far unpublished relevés recorded in the study area and relevés taken from literature sources. The results of statistical analysis show that the marshland vegetation of the Central Balkans is characterized by an exceptional variety of associations. On the basis of differences in the qualitative and quantitative composition of the species, 27 associations were recognized and described, *Typhetum angustifoliae*, *Typhetum latifoliae*, *Typhetum domingensis*, *Phragmitetum australis*, *Schoenoplectetum lacustris*, *Glycerietum maximae*, *Equisetetum limosi*, *Glycerietum fluitantis*, *Glycerietum notatae*, *Sparganietum erecti*, *Oenanthe aquaticae-Rorippetum amphibiae*, *Phalaridetum arundinaceae*, *Eleocharietum palustris*, *Butometum umbellati*, *Bolboschoenetum glauci*, *Caricetum gracillis*, *Caricetum ripariae*, *Caricetum vulpinae*, *Caricetum rostrato-vesicariae*, *Cyperetum longi*, *Caricetum elatae*, *Claditum marisci*, *Caricetum paniculatae*, *Caricetum acutiformis*, *Bolboschoenetum maritimi continentale*, *Schoenoplectetum tabernaemontani* and *Oenanthe fistulosae-Beckmannietum eruciformis*, within 5 orders and 7 alliances in the vegetation class *Phragmito-Magnocaricetea*. Wide distribution and high frequency of occurrence on marshland habitats of the Central Balkans were determined only for three phytocoenoses - *Phragmitetum australis*, *Typhetum latifoliae* and *Sparganietum erecti*.

**KEYWORDS:** marshland vegetation, floristic composition, syntaxonomy, distribution

Poster presentation 54 04 02

### PLANT ASSOCIATIONS OF AQUATIC HABITATS IN AZERBAIJAN

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Azerbaijan is one of the regions rich in plant diversity in the Caucasus region. Water-swamp ecosystems cover large areas in Azerbaijan. In this research, classification and syntaxonomical diversity of plant associations spreading in aquatic habitats of Azerbaijan have been revealed. During 2010-2016, plant samples from these areas were collected. Herbarium specimens are stored in the Azerbaijan MBA Botanical Institute herbarium laboratory. Along the banks of many stagnant coastal waters and lakes, plant assemblages are in the form of stains of different types of vegetation (desert, semi-desert, steppe, meadow, etc.). These troops are in Azerbaijan's order and mountainous areas, Kizilagaç Gulf Yellow Sea, Ağzibirçala, Akgöl, Candar, Büyük Alagöl, Gökgöl, Hacıgabal wide spread around the lake. Different types of vegetation types are found in the form of stains and on the coast of the Caspian Sea. A total of 11.2% of the Azerbaijan flora is present in aquatic habitats: 502 taxa belonging to 62 families and 208 genera. Aquatic vegetation of Azerbaijan includes: vegetation of permanently flooded habitats, coastal water-swamp associations, wet grass associations, aquatic forest associations and hydro-halophyte plants and their associations. The results of these investigations show that in the aquatic ecosystems of Azerbaijan: a total of 75 plant associations belonging to 12 classes, 17 orders and 25 alliances.

**KEYWORDS:** aquatic habitat, forests, diversity, Azerbaijan

Poster presentation 55 04 60

### NEW FLORISTIC AND SYNTAXONOMIC DATA FROM THE RICE FIELDS IN BULGARIA

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The rice is not very important crop in Bulgaria, but the rice fields form temporary aquatic ecosystems. They are highly dynamic and the related to them biotic communities develop rapidly and have very specific flora and vegetation. Until now these rice fields in Bulgaria have not been investigated not only floristically but also syntaxonomically. The aim of this



study is to make an attempt to establish more detail researches on the Bulgarian rice fields and also on their ecological, floristic and dynamic peculiarities. Another goal is to establish the syntaxonomic position of such communities from Bulgaria in the classification of the anthropogenic vegetation in Europe. The floristic and phytocenological studies of the rice fields in Plovdiv and Pazardzhik Districts (Thracian Plain in Central Bulgaria) were carried out in 2016-2017, according to the methods of the sigmatic school. Cluster analysis of the relevés was made using SYNTAXA software. The average linkage method (UPGMA) was applied and floristic similarity among relevés was evaluated according to the Horn's Index. As a result from the studies two new to Bulgarian vascular flora alien freshwater species were found: *Heteranthera reniformis* (Pontederiaceae) and *Rotala ramosior* (Lythraceae). On the basis of several phytocoenological relevés the following new to Bulgaria syntaxa were identified: class *Oryzetea sativae* Miyawaki 1960. The cluster analysis did confirm some high level of floristic and ecological similarities between rice weed communities. Probably they all belong to the widespread in the rice fields in Europe association *Oryzo sativae-Echinocloetum crus-galli* Soo ex Ubrizsy 1948. Typical for this association is the presence of many new exotic weeds like *Ammania coccinea*, *Lindernia dubia*, *Heteranthera rotundifolia* and *H. reniformis*, *Rotala ramosior*, *Diplachne fascicularis*, etc. and naturalized neophytes like *Cyperus difformis*, *Scirpus mucronatus* as well as typical for the Bulgarian flora hygrophytes like *Persicaria lapathifolia*, *Pycreus flavescens*, *Typha angustifolia*. To clarify all floristic and ecologic peculiarities of the rice fields in Bulgaria some new and more detailed floristic and phytocoenological investigations will be necessary which will help also for the clarification of some conservation and economic aspects of rice croplands and for their better management.

**KEYWORDS:** rice fields, aquatic weeds, weeds, alien plants, *Oryzetea sativae*

Poster presentation 56 04 07

### FLORISTIC COMPOSITION OF THE JUNIPERUS COMMUNITIES IN THE TURKISH PART OF THE COLCHIS SECTOR

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*Juniperus* is represented by 11 taxa in Turkey. It is distributed from sea level to 2700 m and it has a very diverse life forms, and high adaptation capacity. *Juniperus* species can form various communities, with different floristical and ecological features. There are floristic studies about the Turkish part of Colchis sector, but only a few of them has given some

information about the floristic composition of the *Juniperus* communities. The aim of the study is to lighten the floristic composition of the *Juniperus* communities of Colchis Sector in Turkish part. In this study, the floristic composition of the *Juniperus* communities of Colchis sector in Turkey determined partly with field surveys, which were periodically done in Rize from June to October between the years 2016-2017, and current literatures, from Trabzon and Artvin. The plant materials were collected with all organs and dried according to standard herbarium rules. The samples were identified with the Flora of Turkey and checked with Turkish Plant List. One sample of each collected plant materials were stocked in the herbarium of Biology Department, Science and Art Faculty, Recep Tayyip Erdoğan University. A total of 307 plant taxa were determined in the *Juniperus* communities of Turkish Colchis sector. 15 taxa, *Abies nordmanniana* subsp. *nordmanniana*, *Acer campestre* var. *campestre*, *A. cappadocicum* var. *cappadocicum*, *A. cappadocicum* var. *stenocarpum*, *A. trautvetteri*, *Picea orientalis*, *Pinus sylvestris*, *Pyrus communis* subsp. *caucasica*, *Quercus petraea* subsp. *iberica*, *Salix caprea*, *Sorbus aucuparia*, *S. caucasica* var. *yaltrikii*, *S. torminalis* var. *torminalis*, *Tilia rubra* subsp. *caucasica*, *Frangula alnus* subsp. *alnus* are three. 20 taxa, *Pyracantha coccinea*, *Rhododendron caucasicum*, *R. luteum*, *R. ponticum*, *Ilex colchica*, *Rhus coriaria*, *Juniperus communis* subsp. *saxatilis*, *J. oxycedrus* subsp. *oxycedrus*, *J. sabina*, *Rosa canina*, *Rubus caucasicus*, *R. discolor*, *R. platyphyllos*, *Vaccinium arctostaphylos*, *V. myrtillus*, *Pyracantha coccinea*, *Cornus mas*, *Corylus avellana* var. *avellana*, *Euonymus latifolius* subsp. *latifolius*, *Empetrum nigrum* subsp. *hermaphroditum* are shrub and a takson, *Hedera colchica*, is a liana. The rest of the total taxa (271) are herbs.

**KEYWORDS:** flora, Colchis sector, *Juniperus* communities, Turkey

Poster presentation 57 04 53

### SYNTAXONOMY OF THERMOPHILOUS FOREST FRINGES (GERANION SANGUINEI TX. IN T.MÜLLER 1962) IN NORTHERN BULGARIA

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Subxerophilous fringe communities in the Southern and Central parts of the Balkan Peninsula belong to the alliance *Lathyro laxiflori-Trifolion velenovskyi* (Čarni et al. 2000) Čarni 2005. However, one also can expect the occurrence of the fringe communities from the alliance *Geranion sanguinei* Tx. in T.Müller 1962 in the Moesian area influenced by continental climate from Pannonian and Pontic regions. The aim of the research is to study the diversity of the marginal forest fringe communities from the class *Trifolio-Geranietae sanguinei* T.

Poster presentation 59 04 28

### PATTERNS OF CHASMOPHYTIC PLANT SPECIES RICHNESS AND THEIR FUNCTIONAL TRAITS ALONG ELEVATIONAL GRADIENTS

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Rocky formations, mainly limestone ones, are shelters of significant, rare and with limited distribution plant taxa, many of which occur exclusively on them. The habitat type 'Calcareous rocky slopes with chasmophytic vegetation' is included in Annex I to Directive 92/43/EEC under the code 8210. Cliffs and slopes with high inclination are usually well preserved and are known as natural refuge for rare plants since they are characterized by a specific floristic composition and they host a large number of endemic species. The aim of this study is to investigate elevation gradients of chasmophytic species (obligate and facultative ones) which occur in high altitudes in Greece combined with their trait based functional diversity and their functional trait distribution. Functional traits influence plants abilities to live in specific habitats and conditions and can influence any plants interactions with the environment. Functional traits can provide us information in order to understand the changes in species richness and composition. For this purpose a database has been created including chasmophytic taxa, their geographical distribution in Greece, their elevation range and their functional traits. The traits included in the database represent vegetative characteristics including longevity, max plant height, life form, growth form, canopy structure, leaf surface texture etc, plant taxa ecological preferences and regenerative characteristics including flowering period start-end, flowering period length, fruit type, flower size etc. In a cliff community the main method of reproduction is seeds as stolons or runners cannot adapt to the ecological conditions. This fact favours the perennials who can establish in cracks and crevices instead of annual plants that are rather rarely encountered on cliffs while trees and shrubs are not frequently occurring on cliffs. Based on the different data included in the database an analysis was carried out concerning distribution patterns of chasmophytic plant species and their functional traits on different altitudinal gradients.

**KEYWORDS:** cliff flora, obligate chasmophyte, distribution patterns, functional traits, Greece

Müller 1962 in Northern Bulgaria (Danubian Plain, Forebalkan, and Northeast Bulgaria) and to try delimitate them off the "pure balkanic" fringe communities. In the region, thermophilous herbaceous fringe communities are associated mainly with open places and edges on the sparse xerothermic oak forests on calcareous hills, and also with complexes of sub-continental steppes and scrubs. The most remarkable species in such communities are *Acanthus hungaricus* (Borbás) Baen., *Cytisus kovacevii* Velen., *Dictamnus albus* L., *Hieracium virosum* Pall., *Paeonia peregrina* Mill., *Rosa gallica* L. etc. A data set of 54 phytosociological relevés made during 2015-2017 was performed and analyzed using Juice 7.0 software package by TWINSPAN protocol. Diagnostic species were chosen on the basis of fidelity measure. Four communities are distinguished, one of them identified as the association of *Galio glauci-Dictamnium albi* van Gils et Kovacs 1977, and three others are preliminary defined as new associations. The classification scheme of the *Geranion sanguinei* communities in northern Bulgaria is discussed and the characteristics of the distinguished syntaxa are presented.

**KEYWORDS:** *Geranion sanguinei*, forest fringe, syntaxonomy, Bulgaria

Poster presentation 58 04 38

### CONTRIBUTION TO THE KNOWLEDGE ON RELIC STIPA SPP. DOMINATED ULTRAMAFIC GRASSLANDS OF THE CENTRAL BALKANS

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The heterogeneity of the ultramafic vegetation of the Central Balkans is rather great. Although this vegetation has been continuously investigated, some gaps are still to be filled. For example, the lack of available data on ultramafic vegetation of the Republic of Macedonia is evident. In that light we have investigated different relic *Stipa* species-dominated dry grasslands over ultramafic bedrock. Twenty relevés were made and compared to the similar vegetation types (i.e. *Potentillion visianii*, *Centaureo kosaninii-Bromion fibrosi*, "*Thymion jankae*", *Saturejo-Thymion* and *Alyssion heldreichii*) by means of numerical analyses. We distinguished one new association and one informal community, providing them with diagnosis and lists of diagnostic, constant and dominant species. We also determined their syntaxonomic positions and relations to ultramafic syntaxa from the neighbouring countries.

**KEYWORDS:** Central Balkans, Macedonia, relics, steppe-like, *Stipa*, ultramafic

Poster presentation 60 04 74

### CONTRIBUTION TO THE SYNECOLOGY OF ENDEMO-RELIC SPECIES *RAMONDA SERBICA* PANČIĆ (GESNERIACEAE)

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As paleoendemic, Tertiary relict and poikilohydric plant the *Ramonda serbica* was a frequent target species in numerous morphological, physiological and phytosociological studies up to now. Nevertheless, some gaps in its synecology has left, especially when we are speaking about its xerophytic and xero-mesophytic plant communities. Over its whole distribution range (Albania, NW Greece, W Macedonia, SW and NE Montenegro, SW, SE and NE Serbia, Kosovo, and NW Bulgaria) the *Ramonda serbica* was found in 14 plant communities, distinguished into 3 ecological groups: xerophytic communities in open gorges under the strong influence of the Mediterranean climate, mesoxerophytic communities in warm and open gorges under the influence of continental climate and higro-mesomorphic communities with mosses on very humid and shady gorges and canyons. Although the mosses are stressed within the third group, they also participate in xerohytic and xero-mesophytic communities, sometimes reaching up to 50% coverage. Due to the fact, an objective of this research is to study the bryophyte component of xerohytic and xero-mesophytic *Ramonda serbica* communities and to test a hypothesis that bryophytes are also a significant component of these plant communities, or even a differential species. The vegetation research has been conducted in Montenegro (ass. *Hieracio-Ramondaetum serbicae* Lakušić 1968, *Geranio-Ramondaetum serbicae* Stevanović & Bulić 1989, *Valeriano-Ramondaetum serbicae* Z. Bulić 1991), and Bulgaria (ass. *Ramonda serbica*- *Festuca dalmatica* Velčev et al. 1973, *Ramonda serbica* - *Sesleria rigida* Velčev et al. 1973, *Ramonda serbica*- *Dianthus petraeus* Velčev et al. 1973, *Ramonda serbica*- *Ceterach officinatum* Velčev et al. 1973) during period from May to July 2018. The relevés have been performed according to the Braun-Blanquet method and stored in the TURBOVEG database. The bryoflora of investigated *Ramonda* communities is represented by ca. 25 species. Xerophytic and more thermophilic communities are characterized by presence of *Homalothecium sericeum*, *Tortula muralis*, and *Preissia quadrata*, while *Neckera crispa*, *Hylocomium splendens* and *Plagiomnium undulatum* are typical for more meso-higrophytic communities. Syntaxonomical diversity of *Ramonda serbica* are still poorly studied in regional aspect.

Future analysis and collecting of new data, including bryophytes, as well as lichens, are necessary for determination of its synecology on Balkan peninsula.

**KEYWORDS:** *Ramonda serbica*, synecology, bryophytes, rocky outcrops

Poster presentation 61 04 33

### HABITAT AND PLANT SPECIES DIVERSITY OF CENTRAL ANATOLIAN SALT LAKES

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Central Anatolia has many salt lakes including the largest salt lake of Turkey "Tuz Lake". These salt lakes are surrounded by salt marshes and salt steppes. Saltmarshes around salt lakes are classified mainly as wet and seasonally dry. Seasonally dry saltmarshes can also be divided into two types according to soluble ion contents; sodium chloride rich and sulphate rich. The dominant vegetation type in Central Anatolia is steppe vegetation and, between steppe and saltmarshes there is a transition vegetation which is called as salt steppe. From lake shores to the steppe vegetation, floristic composition changes according to the soluble ion content and result in special zonation pattern even in small distances. These different habitat types also increase the species diversity of the area and makes the region a hot spot for Turkey. While we compare the habitats according to their species diversity and endemism ratio, Salt steppes are the richest for species diversity and Sulphate rich wet salt marshes are the highest for endemism ratio.

**KEYWORDS:** Tuz Lake, Turkey, habitat classification, plant diversity

Poster presentation 62 04 48

### SYNTAXONOMICAL DIVERSITY OF ELIN PELIN MUNICIPALITY, SOFIA DISTRICT, BULGARIA

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Elin Pelin municipality is situated in western part of the country, closely to Sofia city and covers an area of 433 km<sup>2</sup>. The altitude varies from 500 to 1657 m a.s.l. The area is char-

acterized by a great diversity of abiotic conditions (soil and bedrock types, etc.). The main aim of the study is revealing the syntaxonomical diversity of vegetation as well as assessment of threats for their long term sustainability. During 2017 field working season 303 relevés were collected following the Braun-Blanquet approach. The nomenclature of species was standardized according to Euro+Med PlantBase. This initial dataset was analyzed using EuroVegChecklist Expert System in JUICE programme as well as the Modified TWISPAN were used for determination of vegetation classes. PC-ORD was used for classification below class level - from order to association and community levels. Syntaxonomical diversity is represented by 22 classes (*Carpino-Fagetea sylvaticae*, *Quercetea pubescentis*, *Alnetea glutinosae*, *Salicetea purpureae*, *Robinietae*, *Crataego-Prunetea*, *Lemnetea*, *Potamogetetea*, *Phragmito-Magnocaricetea*, *Isoëto-Nanojuncetea*, *Bidentetea*, *Festuco-Puccinilletea*, *Molinio-Arrhenatheretea*, *Festuco-Brometea*, *Nardetea strictae*, *Koelerio-Corynephoretea canescentis*, *Artemisietea vulgaris*, *Epilobietea angustifolii*, *Papaverietea rhoeadis*, *Digitario sagittalis-Eragrostietae minoris*, *Polygono-Poetea annuae* and *Sisymbrietea*), 29 orders, 39 alliances, 46 associations and 29 communities. Xero-mesophytic and xerophytic woodlands and grasslands have widest distribution on the territory of municipality. Elin Pelin municipality preserves great syntaxonomical and habitat diversity. It is characterized by dominance of xerophytic and xero-mesophytic woodlands, shrubland and grassland vegetation types. Totally 4 orders, 10 alliances и 21 associations were registered for the first time for Bulgaria.

**KEYWORDS:** Braun-Blanquet approach, vegetation classification, Bulgaria, numerical methods

Poster presentation 63 04 06

### DIVERSITY OF HABITAT TYPES AS A RESULT OF TERRAIN PARAMETERS AND ANTHROPOGENIC IMPACT – EXAMPLES FROM THE CITY OF SLATINA, CROATIA

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Habitat mapping has recently become a segment of systematic botanical research in the City of Slatina (Virovitica-Podravina County) undergoing since 2009. The area spreads across 168 km<sup>2</sup> and is geographically located between the Drava River and Papuk Mt, therefore a substantial diversity of habitat types is to be expected. The terrain elevation ranges from 102-262 m, containing both lowland and colline habitats. The produced habitat map, scaled at 1:5000, is mostly the result of physical surveys with over 7500 field observations gathered throughout 2016. A small exception were remote and inaccessible areas that were mapped entirely by photo-interpretation.

The minimal mapping unit for polygons was 250 m<sup>2</sup>. Smaller areas were mapped as point habitats, whereas line vectors were used for roads and streams. Consequently, 596 points, 883 km of lines and 4049 polygons have been created through interpretation of the input data, each part contributing to the distribution of a specific habitat type. Currently unpublished results count a total of 185 habitat types on various classification levels following the National Habitat Classification v.4. Habitat diversity is primarily affected by different terrain parameters such as elevation, aspect and slope, but also soil types and flooding intensity. Additionally, habitats are nowadays frequently shaped by a certain degree of anthropogenic impact. Generally, the whole area is essentially defined by agriculture (42%) and forest/shrub (47%) land cover. The lowland part of Slatina is predominantly covered by *Carpino betuli-Quercetum roboris* forest, whereas the elevated terrain mostly belongs to *Epimedio-Carpinetum betuli* vegetation. Furthermore, woodland stands of invasive *Robinia pseudoacacia* are very frequent throughout the area. Apart from the forest habitats, ecological variables also affect the distribution of various types of grasslands, most commonly *Bromo-Cynosuretum cristati* and *Arrhenatheretum elatioris*. Numerous streams and flooded areas establish aquatic and wetland habitats, many of them rare and endangered such as *Hottonietum palustris* or *Butometum umbellati*. Due to the strong anthropogenic impact, habitat diversity is complemented by a variety of weed and ruderal habitats. Considering the intensity of physical surveys and high level of detail, the habitat map should serve as an improvement to the currently valid national habitat map scaled at 1:25000.

**KEYWORDS:** habitat mapping, national habitat classification, Slatina, Croatia

Poster presentation 64 04 21

### NICHE ANALYSIS OF FOREST ORCHIDS IN WESTERN SERBIA

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The orchid family is one of the largest and most diverse families in the plant kingdom that includes a lot of rare and threatened species. Although it is known that forests represent important ecosystems that host many orchid taxa, ecological research of forest orchids is limited. In this study, factors affecting the occurrence and abundance of orchids in forest habitats in western Serbia (the central Balkan Peninsula) were analyzed. The objectives were to identify the main ecological factors influencing the distribution and abundance of orchids,

to analyze the breadth and marginality of the ecological niche of orchids, and to define ecological groups of orchids. Data concerning habitat type, altitude, inclination, bedrock type, light regime, moisture, soil pH, nitrogen, temperature and geographical coordinates were used as explanatory variables. Niche analysis was carried out using the outlying mean index (OMI) analysis. In addition, a similarity profile analysis (SIMPROF) was applied to classify orchid taxa into meaningful groups and test the significance of these groups. In total, data of 42 orchid taxa from 1091 localities were analyzed. The most species-rich genera were *Epipactis* (with 8 taxa), *Orchis* (6 taxa), *Dactylorhiza* (5 taxa) and *Ophrys* (4 taxa). The results of this study have shown that light regime, temperature, moisture, nitrogen, soil pH and altitude most effectively affect the distribution and abundance of orchid taxa within sampled forests. Furthermore, this study highlights the significance of certain forest types and bedrock types in determining the distribution and abundance of orchids. The taxa-rich forest type is *Fagus* forests with 24 orchid taxa, followed by *Ostrya* forests (23 taxa) and *Quercus* forests (20 taxa). The results have provided the values of habitat specialization for each orchid species, and underlined the specialist (*Epipactis distans*, *E. muelleri*, *E. pontica*, *Epipogium aphyllum*, *Ophrys apifera*, *O. insectifera*) and generalist orchids (*Epipactis microphylla*, *E. helleborine*, *Cephalanthera damasonium*, *C. longifolia*, *Neottia nidus-avis*, *N. ovata*, *Platanthera bifolia*, *P. chlorantha*). In total, six ecological groups of forest orchids were distinguished. Ecological preferences of forest orchids shown in this study can be used as a great background in order to successfully design conservation strategies for these plants.

**KEYWORDS:** Orchidaceae, niche analysis, ecology, forest ecosystems, geological substrates, Serbia

Poster presentation 65 04 43

### ECO-BIOLOGICAL CHARACTERISTICS OF FLORA IN PROTECTED AREA "DABETO" VILLAGE NOVI IZVOR (BULGARIA)

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The present study examines the flora in the protected area (PA) "Dabeto", Novi Izvor village in the region of Plovdiv. Eco-biological characteristic of the vascular plants is made, and the species are categorized by biological groups, by life forms and by floral elements. The plants are classified by ecological groups according their regard towards the water, the light and the heat as a factor. Conclusions for the presence by percentage of medicinal flora in the protected zone are made

The diversity of vascular plants in PA "Dabeto" is studied by systematic observations and collecting of material during the vegetative seasons of 2014-2016. The periodicity of the visits is conformed to the climatic conditions in the region and the determined by their phyto-rhythmic. The earliest aggregates are form the beginning of February 2014, and the latest – from the end of September 2016. The results of research of the vascular plants in PA "Dabeto" shows that there are 160 species plants from 124 genera and 42 families. Among these plants the majority are the perennial herbaceous species, the hemicryptophytes, the species with European and Mediterranean origin, the thermophytes, the mezophytes and the heliophytes.

**KEYWORDS:** Protected area "Dabeto", NATURE 2000, flora, vascular plants

Poster presentation 66 04 70

### FUNCTIONAL DIVERSITY OF SAND AREAS IN SERBIA

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Sand dunes in Serbia are characterized by rich diversity plants and animal species. They are shelter for endangered, endemic and rare plant species. Because of domination of negative anthropogenic influence, balance of living organisms and in such ecosystems is degraded. Basic characteristic of sand dunes is pedological substrate. Wind is deposit and roll sand for a long time, and make a formation of dunes and plains between them. Depending on climatic conditions it was formed specific types of vegetation in this areas. In sand areas in Serbia are different successive types of vegetation are present - from pioneer sand steppes to schrub vegetation and forest communities. This paper analyse three sand areas of Serbia and their typical sand vegetation - *Festucion vaginatae* Soó 1929. Analysis is realised on data from Deliblato, Subotica – Horgos and Ram – Zatonje sandstone. All data are obtained from relevés deposited in phytocoenological database using Turboveg for Windows ver 2.92a software. We have analyzed a database of 110 relevés (with the standard Braun-Blanquet approach), 183 plant species and 13 trait variables. We collected data about functional traits: life form, chorotype, height, vegetative production, plant strategy type; dispersal mode, pollination mode, photosynthetic pathway, leaf and/or stem succulent, hairiness, wax-coating, type of storage organs. Data on the functional diversity of the flora were collected from local Flora, but also from available web databases on the functional diversity of the flora. Trait-species and trait-plot matrices were built. We used hierarchical and non-hierarchical cluster analysis to explore functional strategies of

plants on the sand dune areas of Serbia. We used multivariate analysis to explore the variability of functional traits and the factors that explain it. Sand dune vegetation is very fragile and dynamic. Plant functional traits enabled us to distinct the successive stages between pioneering forms of vegetation on the sand and the steppe formations on stabilized sand mass and the regressive stages induced by human activities. Functional groups are widely used in analyses of different ecosystems. Classifications according to functional traits have been seen as a necessary tool for the simplification of floristic complexity in global vegetation models because they reduce the complexity of ecosystems and, therefore are particularly suitable for studies with diverse species composition.

**KEYWORDS:** functional traits, cluster analysis, *Festucion vaginatae*

Poster presentation 67 04 59

### EARLY-SEASON EFFECTS OF FIRE ON SOIL NUTRIENTS AND UNDER-STORY SPECIES DIVERSITY IN TEAK AND CASHEW PLANTATIONS IN SAVANNA WOODLAND OF NIGERIA

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Fire has evolved inseparable from savanna vegetation of Africa, but its effects on soil chemistry and undergrowth diversity in managed plantations is still subject to intense debate. The study assessed early-season effects of fire on soil chemistry and diversity of under-storey species in teak and cashew plantations in savanna woodland of Nigeria. Data were collected on soil pH, organic carbon/matter, N, P, Na, K, Ca, Mg from top and sub soils in fifteen delineated subplots in the burned and unburned zones of Cashew and Teak plantations after annual wildfire had occurred. Species diversity (H') and similarity in the burned and unburned zones of the plantations were assessed using composition and abundances of under-storey flora two months after a fire event. Canonical correspondence analysis was used to determine the relationship of environmental variables (plantation type, soil fertility and fire) with species composition. Burn incidence significantly improved OC, OM and Ca while reducing total N and Mg in the plantations. Concentrations of soil nutrients varied significantly with plantation type but not with soil depth (except Na). Twenty-eight species occurred in the plantations and were distributed among Rubiaceae (10), Poaceae (5), Asteraceae (5), Malvaceae (3), Euphorbiaceae (1), Lamiaceae (1), Portulacaceae (1), Rubiaceae (1) and Solanaceae (1). Burned Cashew plantation had higher species abundance (4123 plants) but lower diversity (2.810) compared with the unburned plan-

tation. The burned Teak plantation had significantly higher species abundance (3942 plants), richness (23 species) and diversity (2.785) than the unburned Teak. Burned and unburned zones of Cashew and Teak plantations had 90.2% and 86.4% of the species similar. CCA ordination revealed fire event, soil OC and OM concentrations in burned Cashew plantation restricted *Tephrosia pedicellata*, *Desmodium. tortuosum*, *Daniellia oliveri*, *Senna obtusifolia* and *Zornia latifolia* to the site. Presence of *Euphorbia heterophylla*, *Eriosema psoraleoides* and *Crotalaria retusa* in burned Teak plantation is linked to the direct influence of fire on soil Na and Ca concentrations rather than on the species. The study concluded that burning utterly influenced soil fertility but differently affected the diversity of undergrowth flora in the plantations.

**KEYWORDS:** Annual fire, plantation, nutrient, soil, undergrowth, woodland savanna

Poster presentation 68 04 75

### FIRST EXPERIENCE IN USING THE RESEARCH AND USER SUPPORT SERVICE (RUS) IN MAPPING VEGETATION, WATER BODIES AND URBAN AREAS BY SENTINEL-2 SATELLITE IMAGERY

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The European Space Agency (ESA) Sentinel-2 satellites have shown great potential in remote sensing practices. In addition, during September of 2017, The Research and User Support Service (RUS) were launched enabling its users to get access to powerful online virtual machines for processing and analysis of satellite images. This exploratory study presents the results of mapping vegetation, water bodies and urban areas using RUS Service. We acquired four Sentinel-2 L2A Multi-Spectral Images (June, August, November of 2017 and January 2018), with bottom atmospheric correction (BOA) and high-resolution RGB, near infra-red and shortwave infra-red bands (10 m) of Danube's middle flow in Vojvodina province (Serbia). We used Normalized Difference Vegetation Index (NDVI), Normalized Difference Water Index (NDWI) and recently proposed Modified Normalized Difference Water Index (MNDWI) for a vegetation, water body, and urban area detection. Afterward, we calculated a mean pixel value for each one of the indices and used those images as input for an unsupervised classification (K-means). Classification results showed that nearly 25% of pixels are grouped in the same class, based on their mean indices values (NDVI = 0.777; NDWI = -0.769; MNDWI = -0.700) which corresponds to dense vegetation such as forests. Pixels classified as sparse vegetation such as grasslands and senescing crops are grouped within 2 classes (NDVI = 0.583 and NDVI = 0.428) with ap-

proximately 29% and 28% share. Water indices showed similar pixel values for these two classes, but with different percentage share, where the first class is represented with 40% (NDWI) and 43% (MNDWI), second class with 24% (NDWI) and 23% (MNDWI). Water bodies are defined with around 6% (NDVI = -0.887; NDWI = 0.909; MNDWI = 0.766). Pixels classified as urban areas are characterized with 10% (NDVI = 0.201), but water indices showed different percentage share with 6% (NDWI = -0.165) and 5.7% (MNDWI = -0.093). The difference in the percentage of same classified pixels can be explained by diverse spectral bands used for calculating each index and points out the necessity of combining indices for exact mapping of different subjects, and classification accuracy assessment.

**KEYWORDS:** ESA, Sentinel-2 L2A, SNAP Toolbox, multi-spectral remote mapping, vegetation index, water indices

Poster presentation 69 04 66

### MODELLING THE SPATIAL DISTRIBUTION OF BEECH-FIR FORESTS (*ABIETI-FAGENION MOESIACAE*) IN SERBIA

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Beech-fir forests occupy major areas of the mountain region in Serbia. This habitat type is characterized by significant plant species richness and high biodiversity, and present one of the priority types of habitats for conservation under Natura2000 network. As habitat degradation and loss still remains a significant driver of biodiversity loss, it is necessary to establish the spatial distribution of these kind of habitat types in Serbia. This study aims to develop a methodology that identifies the potential spatial distribution of beech-fir forest across Serbia, so that their actual extent can be determined based on the already available data on this habitat type using bioclimatic predictors. A total number of 118 relevés were georeferenced, and further used for predictive modelling. Floristic data were used for taxonomic and biodiversity analyses. The comparison of actual and potential spatial distribution models demonstrate differences in presence of beech-fir forests in western (Tara mountain), central (Kopaonik mountain) and southern (Šara mountain) region of the country. Reviewing the habitat distribution based on the climate data, we concluded that the most limiting factors for the distribution of this habitat type are temperature and precipitation. Taxonomic analysis revealed the presence of 262 species belonging to 165 genera, which represents 21% of the total number of genera recorded in Serbian flora. This research contributes to better understanding and use of geographic information sys-

tems in nature conservation and protection of this important Natura2000 habitat type.

**KEYWORDS:** beech-fir forest, geographic information system, habitat type, Natura2000, spatial modelling, taxonomic analysis

Poster presentation 70 04 55

### GIS DATABASE AND CHECKLIST OF VASCULAR FLORA OF THE REPUBLIC OF SRPSKA

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Regarding the floristic diversity, Bosna and Herzegovina (B&H) with the Republic of Srpska (RS) are amongst the richest areas of Europe. Global standards in nature protection require standardized georeferenced biodiversity databases as a part of integrated information system in order to facilitate information exchange and monitoring of species and habitats. This is also foreseen by national legislation in the sectors of nature protection, forestry etc. However, although the flora of B&H has been studied extensively for the last 150 years, there is still no species check list let alone precise and comprehensive database on species distribution. This was the background for the project *GIS Database of the vascular flora of the Republic of Srpska*, which was completed during 2011 and yielded Web GIS application with the spatial information about vascular plant species of RS, gathered from the largest monograph about B&H flora (Beck-Manageta G. 1903-1983: Flora Bosne i Hercegovine). This project collected 58,026 species occurrences for B&H (20,447 occurrences with around 2330 taxa - species and subspecies level - for the RS). In the second phase of this project that was conducted during 2017–2018, we aimed to collect data from phytosociological relevés (published and unpublished). Around 6000 relevés from B&H were digitized, while 2810 of them were georeferenced and then imported into RDBMS PostgreSQL. This way we acquired additional 98,178 occurrences of plant species (49,390 in RS). Together with the results from the first phase that sums up to 156,204 occurrences (69,837 for RS). Database structure complies with Darwin Core standard. Nomenclature and taxonomy were resolved using the GBIF (Global Biodiversity Information Facility) database. One of the results of the project was the first checklist of vascular flora of the RS. It consists of 2731 taxa at species and subspecies level, classified in 676 genera, 122 families and 45 orders. Our rough assessment is that this number represents not more than 80% of total vascular flora of RS. We also estimate that, from the beginning of the project in 2011, only about 50% of data about vascular plants and their distribution in the RS was prepared and imported into database.

**KEYWORDS:** Bosnia and Herzegovina, distribution, nomenclature, plant species, taxonomy

Poster presentation 71 04 41

### THE ROLE OF GEOGRAPHIC INFORMATION SYSTEMS AND REMOTE SENSING IN PLANT BIODIVERSITY INVENTORY DATABASE ESTABLISHMENT; A NEW METHODOLOGICAL APPROACH

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Establishment of the plant biological diversity inventory database contains important processes such as (1) collection of geographically referenced spatial data, (2) utilizing digital forest stands and vegetation maps, (3) updating produced digital maps, (4) determination of spatial distribution of plant species and plant communities using Geographic Information Systems (GIS) and remote sensing techniques, and (5) entering the data in the attribute tables of the obtained digital maps. The information about the systematic (family, species, subspecies, diversity), plant sociology, endemism, red list categories, BERN and CITES status, economic value, coverage areas are important for the biodiversity inventory database. In order to carry out all these processes in a healthy way, there is a need for complete integration of each process's unique methods. In other words a new methodological approach that integrates all these methods together is needed. In this study, a new methodological approach was proposed for the creation of an appropriate plant biological diversity inventory database for any kind of interrogation and spatial analysis. The proposed methodology provided the integration of methods involving terrestrial sampling studies, plant species identification and systematic studies, plant sociology studies, GIS and remote sensing applications, up-to-date digital map productions and database building. This new methodology is thought to be useful and inspirational to researchers and decision makers.

**KEYWORDS:** GIS, Remote Sensing, plant biological diversity inventory database

Poster presentation 72 04 24

### BIODIVERSITY AND ECOSYSTEM SERVICES IN MOROCCO: CURRENT STATE AND HUMAN DEVELOPMENT

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Morocco is characterized by an important genetic diversity represented by a rich and varied flora with 5211 species and subspecies and many natural ecosystems. Biodiversity and natural ecosystems provide the local population with highly diversified services represented by aromatic and medicinal plants, forage plants, melliferous plants, firewood, lumber, mushrooms, etc. Ecosystem services are currently subject to many pressures: overgrazing and deforestation, climate change, including increased drought, urbanization and forest fire. Conscious of the risks that weigh on biodiversity and ecosystem services, Morocco had made an important effort to reverse the tendencies by developing a consistent biodiversity conservation strategies focused on *in-situ* and *ex-situ* conservation. This presentation will be focused on the current state of biodiversity and ecosystem services and their role for the human development and their decline under the action of different pressures (grazing, timber harvest, harvesting of medicinal and aromatic plants, charcoal making...) while emphasizing efforts constructed by Morocco to conserve and sustainably manage biodiversity and ecosystem services.

**KEYWORDS:** Morocco, biodiversity, ecosystem, services, threats, conservation

Poster presentation 73 04 88

### VEGETATION OF LOW XEROTHERMIC SHRUBS OF ALLIANCE *PRUNION FRUTICOSAE* TÖXEN 1952 IN THE VELIKI RIMSKI ŠANAC (SERBIA)

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The Veliki rimski šanac is a described steppe habitat within an ecological network in Serbia, an important plant area (IPA) and a priority habitat for nature protection (NATURA 2000). It represents a loess embankment and a canal (*šanac*) built for the purpose of defence from *Sarmatae tribes*, overgrown with steppe and forest steppe vegetation, about 1800 years old. It is situated in the south Bačka near Novi Sad and stretches in the northeast-southwest direction, 24 km long (Rimski šančevi-Bačko Gradište: 19°58'16.921"E, 45°24'19.03"N).

The area belongs to Pannonian province within the Pannonian-Vlach subregion, that is to say pontic-southsiberian floristic-vegetational region – vegetation of forest steppe zone (alliance *Festucion rupicolae* Soó 1940, as well as *Aceri tatarico-Quercion* Zóly. et Jakucs, 1957). The predominant type of soil is *černozem*, which indicates the steppe-forest steppe character of the area. During the research of the Veliki rimski šanac locality in 2014, for the needs of the area conservation, plant communities *Prunetum tenellae* de Soó 1951 and *Prunetum fruticosae* Dziubaltowski 1926, within the alliance *Prunetum fruticosae* Tüxen 1952, were described. Phytocoenological records were made per Braun-Blanquet (1964), using GPS, and the areas with plant communities of low thermophilic shrubs covering 6350m<sup>2</sup> or 0.82% of total area of the Veliki rimski šanac were chartered. The community of steppe cherry ass. *Prunetum fruticosae* Dziubaltowski 1926 covers 1.12% of the area, as well as *Prunetum tenellae* de Soó 1951 (0,53%), but they are present on a very small number of localities where they appear within several polygons covering smaller areas. Stands of the communities of low thermophilic shrubs of the alliance *Prunetum fruticosae* Tüxen 1952 grow over the crown of the embankment and the slope towards the canal, as low bushes 0.5-1.5 m high, where predominant species are shrubs *Prunus tenella* and *Prunus fruticosa*. They appear along with steppe vegetation, which caused the occurrence of steppe and forest steppe species: *Koeleria gracilis*, *Agropyrum cristatum*, *Chamaecytisus heuffelii*, *Elymus hispidus*, *Brachypodium pinnatum*, *Centaurea scabiosa*, *Salvia pratensis*, *Stachys recta* and others. Stands of the communities *Prunetum tenellae* and *Prunetum fruticosae* are characterised by a significant number of protected and strictly protected species: *Prunus tenella*, *Prunus fruticosa*, *Sternbergia colchiciflora*, *Carduus hamulosus*, *Agropyron cristatum*.

**KEYWORDS:** vegetation, shrub, Veliki rimski šanac

them, for Bosnia and Herzegovina was previously recorded in karstic field Livanjsko polje. During floristic field investigations in 2017 we found one new locality of *Ophioglossum vulgatum* in Nevesinjsko polje. This rare species grows in Nevesinjsko polje in wet grassland community belonging to *Molinion caeruleae* alliance. Nevesinjsko polje is one of well preserved karstic fields of Bosnia and Herzegovina with many suitable habitats for *Ophioglossum vulgatum*, but it was confirmed only at one site. The number of individuals in population is quite small and is presented by less than 100 individuals. The possible threatening factors include habitat loss due to wetland drainage and overpasturing. We propose further population monitoring and official protection of its characteristic habitats types in Nevesinjsko polje.

**KEYWORDS:** *Ophioglossum vulgatum*, Nevesinjsko polje, karstic fields, flora

Poster presentation 74 04 89

### NEW RECORD OF *OPHIOGLOSSUM VULGATUM* L. (OPHIOGLOSSACEAE) FOR BOSNIA AND HERZEGOVINA

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*Ophioglossum vulgatum* L. (Ophioglossaceae) is rare fern species in the flora of Bosnia and Herzegovina, and since now, it has been known only for three localities. This species predominantly occurs in wet grassland ecosystems, and among

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## CONSERVATION OF PLANT DIVERSITY IN BULGARIA

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The plant diversity in Bulgaria consists of about 4100 species, of which 1/4 are of limited distribution. Approximately 12% of autochthonous species are endemics – 174 species are Bulgarian, and 270 species are Balkan endemics. Biodiversity in Bulgaria is subject to the impact of a wide range of threats resulting from human activity or the natural processes in the ecosystems. Major threats are related to the degradation, fragmentation and loss of habitats as a result of human activity or climate change; overexploitation of natural resources; unregulated felling of forests; fires; air pollution; invasive alien species; genetic erosion, etc. Bulgarian legislation is synchronized with European and world requirements. Bulgaria is a part of a number of conventions and agreements, European directives and regulations, protecting nature and enabling the assessment of its state. Conservation policy is regulated mainly by the Biological Diversity Act which takes care of matters relating to the creation and management of a National Ecological Network, protected species, the species put under the regime of protection and use, *ex situ* conservation of biodiversity, introduction of alien species, and trade in threatened species. The National Ecological Network includes the protected areas designated under Bulgaria's special legislation – Special Areas of Conservation (SAC) and Special Protection Areas (SPAs). According to the Protected Areas Act, six categories of protected areas in line with the norms of the International Union for Conservation of Nature (IUCN) exist in Bulgaria: reserves and managed reserves (90), national parks (3), nature parks (11), natural monuments (more than 350), and protected sites (more than 500). They cover more than 5% of the territory of the country while the Bulgarian part of European Ecological Network Natura 2000 accounts for 34.4% of the territory. In the last few years, the concept of Micro Reserves has been implemented in Bulgaria. It was applied for the first time in Valencia (Spain) and aimed at the conservation of small and isolated populations of rare plant species. In the course of four years, a network of 58 small protected territories has been created, in the 'protected site' category, for conservation of isolated populations of 47 species in the Bulgarian flora. At present, scientists, students, NGOs implement a number of initiatives and activities aimed at improving the status of populations of rare plant species and their habitats.

**KEYWORDS:** Bulgaria, plant diversity and conservation, protected plant species and territories

## INVASIVE ALIEN SPECIES OF VASCULAR PLANTS IN THE BALKAN PENINSULA – CURRENT CONDITION AND CHALLENGES IN RESEARCH

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Balkan Peninsula is one of the World's hotspots of vascular plant diversity which is of particular interest for research and conservation. Invasive alien species are recognized as one of the key threats to biodiversity worldwide. Therefore, they have drawn the attention of botanists in the Balkan countries during the past two decades. The objective of this presentation is to review the most important recent and current projects related to invasive alien plants in the Balkan countries and to outline the main results and the future challenges in research of these species. The analysis is based on published articles, web-sites of relevant projects and the personal experience of the authors. It can be inferred that most studies have been focused on the inventory of the alien flora of the respective country and classification of the species according to their invasive behavior and potential. Numerous new records of alien species for the Balkan countries, lists of the invasive species, and for some countries – lists of all alien species have already been published. Although the lists of invasive alien plants have been based mainly on expert opinion, the most widespread species and these with the strongest impact on the native biodiversity and on the human health and economy in the region can be identified: *Acer negundo*, *Ailanthus altissima*, *Ambrosia artemisiifolia*, *Amorpha fruticosa*, *Cuscuta campestris*, *Erigeron annuus*, *E. canadensis*, *Fallopia xbohemica*/*F. japonica*, *Helianthus tuberosus*, *Paspalum distichum*, *Robinia pseudoacacia*, *Solanum elaeagnifolium*, *Solidago canadensis*/*S. gigantea*, etc. The challenges in research of alien and invasive alien species will be outlined, especially with regard to the compilation of a list of alien plants for the entire Balkan Peninsula, objective assessment of the invasiveness of the species, management of the aliens and the implementation of international binding agreements and regulations, including Regulation (EU) 1143/2014 on the prevention and management of the introduction and spread of invasive alien species.

**KEYWORDS:** aliens, Balkan Peninsula, flora of the Balkan Peninsula, invasive plants, South East Europe

## ASSESSING IMPACTS OF CLIMATE CHANGE ON A CRITICALLY ENDANGERED ENDEMIC SPECIES: *CAMPANULA YALTIRIKII* H. DUMAN

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Climate change is an important cause for extinction of the species. Increasingly rapid warming is reported to occur recently. Endangered species are fragile to climatic changes. Understanding response of the threatened species to climate change is of great importance for conservation planning of biodiversity. Ecological niche modelling (ENM) provides information to assess effects of environmental and climatic conditions on species distribution. The purpose of this study was to predict impact of climate change on a critically endangered species *Campanula yaltirikii* H. Duman. It is a critically endangered endemic species which occurs in cracks of calcareous rocks in a narrow range of distribution. The species is also threatened by overgrazing by goats of the local shepherds. Current and future potential distributions of *C. yaltirikii* were analyzed and based on results future distribution predicted with ENM, by using Maxent (Maximum Entropy) software. MIROC5 (Model for Interdisciplinary Research on Climate) climate change scenario for the year 2070, which was created based on fifth IPCC report, was used for projecting future potential distribution of the species. A total of 27 GPS records of the species' localities were obtained from field works. Including edaphic and topographic factors 15 environmental variables and 19 climatic variables used as predictors. SRTM digital elevation model used with 30 m resolution to create topographic variables. The Jackknife evaluation results indicated geological formation, soil groups and elevation as main factors influencing *C. yaltirikii*'s distribution. Additionally, analysis of variable contributions results showed that max temperature of warmest week (b5) and mean diurnal temperature range (mc2) are the most effective climatic variables for current and future model, respectively. To conclude the model indicated that *C. yaltirikii* has really narrow suitable habitat currently. Potential distribution area for the species will probably shrink by 2070, if the climate change prediction model is correct. Therefore, the species needs rapid conservation measures.

**KEYWORDS:** global warming, species distribution modeling, conservation biology

## CONSERVATION ACTIVITIES FOR A POPULATION OF *DACTYLORHIZA KALOPISSII* – A GLOBALLY THREATENED SPECIES

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The Balkan sub endemic species *Dactylorhiza kalopissii* is a globally endangered, included in Annex IIb of the Directive 92/43 EEC. The distribution in Bulgaria is a local one, in vulnerable habitats – alkaline fens and wet meadows. We discovered the population in Sinite Kamani Natural Park in 2008, the number of individuals was 30 in 2 closely situated areas. The suitable habitats are extremely limited there and the area of the population is less than 0.05 ha. The monitoring during the period 2009-2013 showed a 50% decline of the population and no recruitment. The management of the habitat and support activities for the recruitment of the plants were done in 2013-2014 as a part of a conservation project with EU financial support. The decisions about the methods and seasons of activities are discussed and described. A mowing of grass cover and clearing of shrubs were done during the autumns, in both years, for the effective and potential area of the population as well as for a buffer belt around. Precise data about population number, phenology, flowering and fruiting were regularly collected. For the assisted reproduction, small spots (5-10 cm<sup>2</sup>) were cleared of plant cover and tiny seeds sown in September 2013, and August and September 2014. We used no more than 15 % of the seed pots of an individual for this. The single new individuals at marked spots (where seeds were sown), appeared at 2015 and more abundantly in 2016. Further monitoring revealed a significant increasing of the numbers of plants and self-recruitment in other parts of the appropriate habitat. In short, 5 years, term, the results are positive. We recommend a regular monitoring and, considering the small area of the population, the lack of appropriate habitats and the biology of the species, periodical conservation activities.

**KEYWORDS:** *Dactylorhiza kalopissii*, orchid population studies, conservation activities

Oral presentation 05 05 06

### RARE PLANT SPECIES ON THE SANDY SHORE FROM DANUBE DELTA, ROMANIA: THREATS AND CONSERVATIVE STATUS

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Along the Danube Delta coastline, between the two mouths of the River Danube, Sulina and Saint George, it has been developed a typical beach-fore dune system. This sandy shore section belongs to the marine delta plain that is a wilderness area *sensu* World Conservation Union. Due to geographical position, particular climate and substrate, this area harbors high plant species diversity, rare taxa and habitats. This research was focused on identifying plant species, threatened taxa with different conservational status and current main threats within the beach-fore dune system, the most sensitive and vulnerable area of the coastal region. The floristic survey was carried out using the transect method, perpendicular to the shoreline following each morphological feature of the beach-fore-dune system. Species were identified by referring to standard floras and floristic keys. The conservative status of each taxon was established according to the national Red Book of high plants and annexes of the Council Directive 92/43/EEC on the Conservation of natural habitats and of wild fauna and flora. Our results show occurrence of 38 taxa that belong to 15 families, typical and non-typical for the sandy shores from temperate regions. Floristic spectrum shows a mixture of elements but prevalent are Pontic and Ponto-Caspian elements. In the case of the regional endemism *Convolvulus persicus* L., the study area is an ecological refuge taking into account that the area covered by this psammophilous plant species has been increased in the recent years. Eleven plant species are endangered and vulnerable. The main threats of shore taxa are both natural and anthropogenic: habitat loss and destruction caused by sea level rise, shore erosion, invasive species, and touristic activities. In order to preserve rare plant species and their associated habitats from this area, special conservation management strategy needs to be applied.

**KEYWORDS:** beach fore-dune system, floristic spectrum, regional endemism, ecological refuge, *Convolvulus persicus* L., habitat loss

Oral presentation 06 05 11

### SOME NOTES ABOUT PROTECTED BRYOPHYTA IN MONTENEGRO

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According to the current legislation (Official Gazette of Republic of Montenegro, N° 76/06, 12 December 2006), 26 species of bryophyta are protected in Montenegro, which comprise 23 mosses: *Brachythecium geheebii*, *Campyliadelphus chrysophyllus*, *Hypnum fertile*, *Pseudoleskea saviana*, *Homalia webbiana*, *Neckera pennata*, *Buxbaumia viridis*, *Dicranum viride*, *Paraleucobryum sauteri*, *Fissidens curvatus*, *Ephemerum recurvifolium*, *Ephemerum sessile*, *Funaria microstoma*, *Funariella curviseta*, *Grimmia caespiticia*, *Grimmia fuscolutea*, *Orthotrichum patens*, *Molendoa hornschurchiana*, *Tortula lingulata*, *Syntrichia princeps*, *Weissia levieri*, *Trichostomum triumphans*, *Ulota crispa* and 3 liverworts: *Mannia triandra*, *Cephaloziella calyculata* and *Lophozia ascendens*. Bryological research on the territory of Montenegro had been intensified during the last decade, with an emphasis to the areas without previously known or obsolete data (e.g. Boka Kotorska Bay, Orjen, Rumija, Bjelasica, Komovi and Durmitor mountains) resulting in the increase of the number of known species to almost 700. This paper synthesizes all published data on the protected bryophyta species as well as previously unpublished data in Montenegro in order to provide recommendations on the further treatment of these taxa, as well as the proposal for the protection of new species according to their status (IUCN criteria, Bryophytes Red List in Europe).

**KEYWORDS:** Bryophyta, protected species, Montenegro

Oral presentation 07 05 22

### AN OVERVIEW OF ALIEN AQUATIC FLORA OF CROATIA – NEW INSIGHTS FROM NATIONAL MONITORING OF MACROPHYTES

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Invasions of alien organisms are nowadays considered one of the major threats to biodiversity. Intrusions of alien species in aquatic ecosystems are particularly interesting and heavily

studied, due to the increased spreading potential of organisms in systems interconnected with flowing waters. Data on the distribution of alien aquatic plant species were gathered within the monitoring of surface waters, conducted in order to meet the requirements of the Water Framework Directive. Monitoring of macrophyte vegetation was carried out during the vegetation seasons 2013-2017 in over 600 locations, ultimately covering most of the Croatian territory, being the first comprehensive study of macrophytes in Croatia so far. Watercourses were surveyed along 100 m-long transects, while multiple 6 x 100 m-transects were used when surveying the lakes. Cover and abundance of each macrophyte species were estimated according to Braun-Blanquet scale. We have found altogether five alien species during our research. Three species were recorded only once, in continental part of Croatia: *Elodea nuttallii* (Planch.) H.St.John, *Azolla filiculoides* Lam. and *Lemna minuta* Kunth. Noteworthy, *L. minuta* was not previously recorded in Croatia, therefore this finding (Čarna River) is the first known site. Another newly found species is *Egeria densa* Planch. recorded on five sites in the Neretva River Basin. Furthermore, we provide updated distribution of *Elodea canadensis* Michx. with as many as 19 new records. It was mostly found in the continental part of Croatia, but also in the Cetina River Basin, which is the first record in the Mediterranean part of the country. Along with the data on the distribution, we present the data on habitat characteristics of finding sites, as well as our remarks on the observed trend of its spread. Including the literature data, we can account for nine alien aquatic plants in Croatian waters so far, out of which only *E. canadensis* is considered invasive. Our results show that the number of alien aquatic species in Croatia is still relatively low, and their distribution is rather limited, especially in comparison with Central European countries.

**KEYWORDS:** invasive alien species, allochthonous plants, macrophytes, monitoring, Croatia, Water Framework Directive

Oral presentation 08 05 16

### RIVERS IN SERBIA AS IMPORTANT PLANT INVASION CORRIDORS

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The spread of invasive alien species (IAS) has become one of the burning research topics over the past few decades. Invasion corridors, as pathways enabling species dispersal over long distances, are crucial in understanding invasion dynamics. Since rivers as invasion corridors have not previously been studied in our country, the aim was to determine which

river catchments are most affected by the spread of invasive alien plant species in Serbia. Over the period of four years (2013-2016) field research was carried out during the summer months, along the course of 36 rivers. The total number of field sites was 228, where the presence and abundance of invasive alien plants was recorded along 100 m long transects. Seven economically important IAS were chosen for the analysis. Of the selected seven species, *Robinia pseudoacacia* and *Xanthium strumarium* were recorded along the course of 32 rivers (88.89% of rivers analysed), at 145 and 105 field sites, respectively. The most invaded catchment areas by *R. pseudoacacia* were those of Danube (83.33% field sites), Zapadna Morava (68%), Južna Morava (62.07%) and Timok (60%) rivers. Conversely, the most affected catchment areas by the presence of *X. strumarium* were those of Kolubara, Sava and Timok rivers, with 80%, 76.92% and 73.33% of field sites invaded, respectively. While *Conyza canadensis* was predominantly found along the rivers belonging to Timok (73.33%), Južna Morava (59.26%) and Kolubara (45%) catchments, *Echinochloa crus-galli* and *Amorpha fruticosa* were documented mostly within the rivers belonging to the catchment areas of Sava (69.23% and 76.92%, respectively) and Danube (50% and 56.67%) rivers, with the addition of Zapadna Morava (64%) for *E. crus-galli* and Timok (66.67%) river catchments for *A. fruticosa*. *Ambrosia artemisiifolia* was mostly documented along the rivers belonging to Sava (76.92%) and Kolubara (70%) catchment areas, while *Echinocystis lobata* was predominantly found in the catchment areas of the Timok, Velika and Zapadna Morava rivers, with 66.67%, 56.25% and 52% of field sites, respectively. In conclusion, it can be said that the catchment areas of the Sava, Timok and Zapadna Morava rivers are summarily the most affected by the invasion of these seven chosen IAS.

**ACKNOWLEDGMENTS:** The authors would like to acknowledge the support of the Ministry of Education, Science and Technological Development of the Republic of Serbia (Projects No. TR31018, TR31043, III43002).

**KEYWORDS:** invasive alien plants, invasion corridor, river, catchment area, Serbia

Oral presentation 09 05 31

### FLORA OF TRAFFIC CORRIDORS OF THE CITY OF SARAJEVO

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The earliest data on the flora of Sarajevo dates back to first decades of floristic exploration of this region, explicitly to 1882. The most important historical contribution to the knowledge of Sarajevo flora consists of a series of papers written by Karlo

Maly during the 1904-1940 period. Other authors have mentioned interesting, weedy or new plants found in the area, but only occasionally. Until mid-1990s, this area was almost completely neglected by botanists. Only in the last two decades several papers, mostly focused on alien plants, appeared. The systematic mapping of urban flora of the city of Sarajevo has been conducted for the first time since summer 2015, on an area of 32 km<sup>2</sup>. Unfortunately, scarcity of old data does not allow relevant comparisons with recent flora, because of uneven temporal distribution and spatial inaccuracy of data. Here we analyze the flora of traffic corridors of the city of Sarajevo as partial result of the above-mentioned mapping. The research was focused on following habitat types: (1) promenades, used by pedestrians and bicycles, (2) avenues, intended for vehicles and lined with planted trees, (3) roads and roadsides without trees, (4) green patches along the tramlines and (5) railways. So far, around 15.000 records have been gathered for the flora of Sarajevo, of which around 4.500 in discussed habitat types. The total number of corridor taxa (398) represents about two thirds of Sarajevo's total flora, which indicates the general importance of corridors for the diversity of urban vascular plant flora. The presence of alien plants indicates that traffic corridors for vehicles are more susceptible to disturbance and harbour more alien, naturalized and invasive species than corridors for pedestrians. The site specificity of railway and tramway grounds favors the appearance and spread of many interesting taxa, including halophylous *Puccinellia distans*, neophytic *Amaranthus albus*, *Panicum capillare* and *Euphorbia prostrata* from North America and *Senecio inaequidens* from southern Africa, which were not found in other investigated habitat types. Analyses of life forms, origin and time of first record in the area, and comparison between mentioned habitat types and complete flora of Sarajevo will be presented.

**KEYWORDS:** Sarajevo, urban flora, traffic corridors, alien plants

Poster presentation 10 05 32

### IDENTIFYING NATURA 2000 HABITATS IN THE WATERSHED OF THE MIDDLE SECTION OF DEVOLL RIVER, SE ALBANIA

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The watershed of the middle section of Devoll River is located between the districts of Korça, Pogradeci and Gramshi. The geographical location of this area, complex topography, climate characteristics, different rock substrata and soil types as well as hydrological conditions, are factors contributing to a rich flora and diversity of natural habitats. Identification and mapping of Natura 2000 habitats types within the study area,

basing mostly in identification of their typical plants species, was the main goal of the study. It was carried out during the period 2016-2018. The habitats mapping was done by using Geographical Information Software (GIS), version 10.1. As result, the most representative habitats are: 9530\* (Sub-) Mediterranean pine forests with endemic black pines; 91M0 Pannonian-Balkan turkey oak –sessile oak forests; 91L0 Illyrian oak-hornbeam forests (*Erythronio-Carpinion*); 9250 *Quercus trojana* woods; 9340 *Quercus ilex* and *Quercus rotundifolia* forests etc. This area supports even a wide range of semi-natural and modified habitats, especially those intensively used for firewood collection and grazing that constitute the majority of these habitats, while artificial habitats represented mostly by agricultural land and pine plantations on eroded slopes that cover small sections of the study area.

**KEYWORDS:** watershed of the middle section of Devoll River, Natura 2000 habitats, GIS.

Poster presentation 11 05 33

### NATURA 2000 HABITATS MAP OF "DIVJAKE-KARAVASTA" NATIONAL PARKIN ALBANIA

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The wetland ecosystem of "Divjake-Karavsta", is located in Lushnje city, in center part of Albania. It is a National Park, II category since 2007. The aim of this paper is to study the habitats according to Directive of Habitats, mostly by carrying out vegetation's relevees. Based on the local orthophotos and observations during field excursions, a habitat map was elaborated. From a geological point of view the park is composed by sandy and sub sandy sea Quaternary deposits. Due to favorable climatic and edaphic conditions in this park a variety of habitats and endemic species are found. In the park are present *Galatella albanica*, *Orchis albanica* Goelz et Reinhard, which are endemic species of Albania and 3priority habitats such as: Coastal dunes, with the code 1150, Coastal dunes with *Juniperus* spp., with the code 2250, and Wooded dunes with *Pinus pinea* and/or *Pinus pinaster*, with the code \*2270. Such analysis was carried out in frame work of Natura 2000 Project in Albania, which will serve in the future for the Annex I of the Habitat Directive.

**KEYWORDS:** wetland ecosystem, Natura 2000, directive of habitats, ArcMap

Poster presentation 12 05 34

### THE DISTRIBUTION AND STRUCTURE OF THE PLANT COMMUNITIES FOUND IN THE JGHEABURI FOREST OF THE GOVORA RIVER BASIN, ROMANIA

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The thematic area provided in this paper is situated in the Govora river basin, Vâlcea County. Within this study had been aimed the identification, description, diversity, ecological analysis and monitoring of the herbaceous and wooden plant communities, which belong to the Natura 2000 habitats and implicitly of the rare plant species, vulnerable, endemic whitin Jgeaburi Forest from the Govora river basin. In this area we identified the following plant communities: *Hieracio rotundati-Fagetum* Vida 1963) Täuber 1987 (syn.: *Deschampsio flexuosae-Fagetum* Soó 1962; *Stellario nemorumAlnetum glutinosae* (Kästner 1938) Lohmeyer 1957; *Quercetum petraeae-cerris* Sóo (1957) 1969; *Aremonio-Quercetum petraeae* Hoborka 1980 and *Carpino-Fagetum sylvaticae* Paucă 1941. According to the targets of this research, a very important place we gave to the complex study of the habitats: 91M0 Pannonian-Balkan turkey oak sessile oak forests ( CLAS. PAL.: 41.76); 9110 *Luzulo-Fagetum* beech forests (CLAS. PAL.: 41.11.); 91E0\* Alluvial forests with *Alnus glutinosa* and *Fraxinus excelsior* (*Alno-Padion*, *Alnion incanae*, *Salicion albae*)( CLAS. PAL.: 44.3, 44.2 și 44.13.) and 9130 *Asperulo-Fagetum* beech forests (CLAS. PAL.: 41.13. Considering the place where the study had been located to, the eco-pedo-climatic conditions and the anthropic term exerted I have considered that is necessary to develop some ecological studies (and using statistical methods (UPGMA si WPGMA, STYN-TAX 2000) for the plants communities from this area.

**KEYWORDS:** Govora river, plant communities, forests, diversity, habitats

Poster presentation 13 05 35

### PROKLETIJE – NEW NATIONAL PARK IN MONTENEGRO

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A natural sight of outstanding universal value, Prokletije (Montenegro) was inscribed on the national park in 2009. New national park Prokletije in Montenegro is characterized

by distinctive relief, specific hydrography and interesting living world. This paper contains basic physically - geographical characteristics and overall natural and cultural bases of the National Park „Montenegrin Prokletije“ representing one of the most important natural and cultural potentials not only of Montenegro but of encirclement. Considering all previous natural and cultural potentials, this part of Prokletije belonging to Montenegro as well as in surrounding countries where this mountain massif is extended, deserves to be national park. Basic assumptions of historically – natural, physically – geographical, cultural, monumental, biodiversity and other characters are represented here. All these features nominate this area to be, not only national park, but area to be registered in the list of world-natural and cultural heritage.

**KEYWORDS:** National park, protection of nature, sustainable development, Prokletije, Montenegro, Balkan Peninsula

Poster presentation 14 05 02

### METHODOLOGY FOR DETERMINING THE CONSERVATION VALUE OF HALOPHILOUS HABITATS BASED ON FLORISTIC AND VEGETATION PARAMETERS: A CASE STUDY FROM CROATIA

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Halophilous habitats on the Croatian coast of the Adriatic Sea are rare and confined to the narrow strip of the coast that is threatened by urbanization and tourism. To prioritize conservation and restoration actions the conservation value of halophilous habitats should be assessed using a simple and replicable methodology which quantifies their value. Here we present the application of such a methodological approach on a small salt lake in Dalmatia (Croatia), which has its shores covered by different halophilous habitats. The current state of these rare habitats is heavily influenced by past use of the area and their persistence is still threatened by the expansion of the small coastal village. To determine the conservation value, the status and the quality of halophilous habitats, we evaluated all the patches with halophilous vegetation along the shore of the lake. The patches represent GIS polygons of uneven size and shape. The evaluation method was based on 10 criteria that reflect the conservation objectives, with the most important criteria being the dominance of diagnostic species and life form spectra (determining the vegetation structure) and the presence of anthropogenic pressure. The scores for the patches were grouped in three classes: Favorable, Intermediate, and Unfavorable; with the highest scores indicating the best conservation value. Patches of the highest conservation value are found on low laying shore in the northern and western part of the lake, while the other halophilous habitats show various degrees of degradation, and thus a lower conservation value.



Overall, the results show that the conservation value of halophilous habitats of this small salt lake is adequate, as most of the patches received an intermediate score, while only 31.7% received the high score. The degradation is most pronounced for the patches disconnected from the lake by anthropogenic features such as an asphalt road and a stone wall. This evaluation method enables precise identification of most valuable patches for conservation and quantifies the overall conservation value of habitats in a particular area. We are confident that this methodology can be a valuable tool for vegetation conservation and habitat quality assessment.

**KEYWORDS:** halophilous habitat, habitat quality, conservation value, evaluation, DAFOR scale, GIS analysis

Poster presentation 15 05 05

**THE PRESENCE OF AN ENDANGERED HABITAT AND THE NECESSITY FOR REASSESSING THE FIRST ZONE OF PROTECTED AREA IN LUBOTEN, SHARRI NATIONAL PARK, KOSOVO\* (according to UNSC Resolution 1244)**

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With anthropogenic pressures growing towards natural habitats, in particular via exploitation of water sources and intensive grazing, a growing threat to areas designed to protect habitats with high biodiversity has been noticed. In order for us to assess the present level of threat, we have studied the correlation between the factual situation of natural habitats and the boundary of the first zone of protected area (IPA). We have made 25 phytocoenological relevés in the studied site, we have recorded the habitat types (based on EUNIS habitat classification and the EU Habitats Directive) as well as took note on present endangered and rare plant taxa. We have noticed that within the massif of Luboten, Sharri National Park, an endangered natural habitat of subalpine moist tall herbs is not covered within the IPA. E5.5723 - Moesian hogweed tall herb communities with *Cirsium appendiculatum* Griseb., as the most distinctive plant species, are tall herb formations of the Balkan Range that are known to harbor several endemic and rare plant species like: *Alchemilla plicatula* Walters., *Trollius europaeus* L., *Pinguicula balcanica* Casper, etc. To further add conservation importance, in these habitats with narrow distribution and fragile environment of fresh water springs (at altitudes of above 1890 m a.s.l.) there is one South-European Orophilous plant species (*Willemetia stipitata* (Jacq.) Dalla Torre) present with the only known locality so far for Kosovo and lives in these fragile and disturbed environments.

The corresponding recorded plant association *Doronico gigantei-Cirsietum appendiculati* Horvat ex Quezel 1969, develops on narrow and fragile habitats along a water stream, on the western slope of the massif. Based on the obtained data on the situation of this habitat, recorded phytosociological relevés and potential threats, we highly suggest extending the first zone for 0.56 km<sup>2</sup> into the northwestern direction of the western slope. Furthermore we suggest that inclusion of this site within the IPA is of crucial importance in order to avoid cascade of natural changes and large scale declines of rare plant communities, plant taxa and their corresponding natural habitats.

**KEYWORDS:** EUNIS, habitat disturbance, Kosovo\*, phytosociology, nature conservation

Poster presentation 16 05 09

**PENETRATION OF NON-PSAMMOPHYTES IN SAND DUNE HABITATS - A STUDY FROM THE BULGARIAN BLACK SEA COAST**

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Bulgarian coastal dunes are under increasing tourist pressure in the past 20 years. Typical psammophytic plants suffer from direct destruction, limitation of available space and pollution. Another threat is the penetration of alien and native but not typical plants in the dune systems. Limited number of studies deal with this problem, but this issue is important because establishment of non-psammophytes provides evidence of changes in the ecosystems structure and function. This study aims to identify how far the different dune types at the Bulgarian Black Sea Coast are affected by non-psammophytes in order to take measures for further conservation. We studied embryonic, white and grey dunes during July – August 2017. Vegetation was sampled at 12 dune complexes within 158 plots along orthogonal to the seashore transects. All vascular plants and their percentage cover as well as the total vegetation cover, and separately the cover of vascular plants and lichens and bryophytes were registered within each plot. The overall species list was additionally separated to five groups: psammophytes, weeds and ruderals, plants typical for grasslands and shrublands, plants typical for forests and alien (including invasive) species. We used analysis of similarities (ANOSIM) to prove differences in species composition in the different dune types and MDS (non-metric multidimensional scaling) to visualize the distances between plots. Differences in number and cover of defined species groups among the three dune habitats were tested with ANOVAs and Tukey's HSD post hoc test was used to identify significant differences. A total 275 plant species were registered: psammophytes – 51, weeds and ruderals – 41, plants typical for grasslands and shrublands –

143, plants typical for forests – 21, alien (including invasive) species – 19, bryophytes – 12 and lichens – 6. Grey dunes contain the highest percentage of non-psammophytic plants, which decrease toward white and embryonic dunes. The native plants typical for surrounding grasslands and shrublands are to be considered as main contributors to the current state, while neophytes are still at low abundance. The studied coastal area is still less influenced by non-psammophytes. The high floristic and habitat diversity deserve further effective protection.

**KEYWORDS:** coastal dunes, plant species groups (alien, forest, grass- and shrubland, psammophyte, ruderal and weed plants)

Poster presentation 17 05 36

**CONSERVATION STATUS OF PSAMMOPHYTIC AND HALOPHYTIC HABITATS AND PLANT COMMUNITIES IN THE COASTAL AREA OF THE DANUBE DELTA**

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The Danube Delta, the third largest river delta of Europe is a complex of natural habitats with a remarkable phytodiversity (2,383 plant taxa). The entire northern coast of Romania is included in the Biosphere Reserve and it is generally well preserved. On the wide sandy beaches and neighboring moist and salty areas, as well as on the sandbanks from the south of the Danube Delta there are many habitats with conservation significance. Some psammophytic and halophytic habitat types protected at European level have a specific structure and floristic composition due to typical physico-geographical and climatic conditions. The sandbars with shifting and stabilized sand dunes, the moist and salty interdune areas, the salty lakes and swampy areas from the proximity of the beaches, as well as the high air humidity and high temperatures in the summer period make the coastal area of the Danube Delta an unique complex of biotopes with psammophytic, halophytic, hygrophytic and steppe plant communities. Appropriate preservation of these habitats is very important at national level because on the southern coast of Romania, which is well developed economically, the natural vegetation of the beaches is very fragmented, present only on small areas and highly affected by human activities (mainly tourism, harbor facilities, etc). In the Danube Delta Biosphere Reserve, human pressure upon the natural habitats and vegetation is high only around some small villages and cities (mainly grazing and recreational activities on the beaches). Based on field observations, this paper will give a description of the particularities and current conservation status of some psammophytic and halophytic

habitats and plant communities of the Danube Delta, of the vulnerabilities and future tendencies of the habitats, and will make proposals for a better management of the coastal area of the Biosphere Reserve. Reducing the anthropogenic pressures upon natural habitats is essential for the adequate preservation of the rare plants and plant communities of the coastal area of the Danube Delta Biosphere Reserve, mainly of the dune vegetation and of the halophytic vegetation, which are rich in rare plant species at national and European level.

**KEYWORDS:** coastal habitats, plant communities, Danube Delta

Poster presentation 18 05 18

**PRELIMINARY RESULTS OF EX-SITU CONSERVATION STUDIES OF ASTRAGALUS BEYPAZARICUS**

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*Astragalus beypazaricus* Podlech & Aytac is a perennial narrow endemic species restricted with gypsaceous soils at Beypazarı (Ankara/Turkey). According to the field researches and IUCN criteria, it is included in CR (Critically Endangered) and is under protection. Although there is an increasing attempt to in-situ protection of this species invasion of distribution area with agricultural fields ex-situ conservation became a requirement. It is aimed to increase the knowledge about ecology and reproduction of this species to support the in-situ conservation studies. 16 individuals from Beypazarı were transferred to the pre-set area with gypsaceous soil at Ankara University campus in 2017. 9 of these 16 individuals survived at their new habitat, 5 of them gave flowers but only 1 fruit was obtained. Under laboratory conditions, new individuals were cultivated from the seeds collected from natural distribution area and planted to the conservation garden in the place of un-survived individuals. During the vegetation period of 2018, 8 of mature individuals bloomed and gave fruits. These seeds are going to be used in production of next individuals. Also the seeds in the soil seed bank germinated and 24 of new individuals started to grow. According to the current observations this species has at least short term persistent seed bank which can be important for conservation studies.

**KEYWORDS:** *Astragalus beypazaricus*, ex-situ conservation, Ankara, endemic, seed bank

Poster presentation 19 05 21

### CONSERVATION STATUS, DISTRIBUTION AREA AND CURRENT POPULATION SIZE OF *CAMPANULA DAMBOLDTIANA*

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*Campanula damboldtiana* P.H.Davis & Sorger is a local, edaphic endemic species which is threatened and narrowly distributed with 3 localities from Ayaş (Aysantbeli), Sincan and Kazan district in Ankara province. *C. damboldtiana* is involved in Bern Convention Appendix-I under the title of "Strictly Protected Flora Species". The goal of this study was to determine the current population size, distribution area of the species and identify main threats on it and re-evaluate IUCN category of *C.damboldtiana*. Between 2016 and 2017 distribution area, population size and threat factors were investigated by field studies. Threat category of the species was re-evaluated by using data derived from the study on the basis of IUCN Red List Categories and Criteria. The number of location is found as 3, total number of individuals is found as 8948, area of occupancy is found as 12 km<sup>2</sup> and extent of occurrence is found as 58 km<sup>2</sup> for *C.damboldtiana*. Also expansion of agricultural fields, soda production plant and excavation works to provide raw material such as clay and marl are being serious threatening factors to this local endemic species. Considering the results of this study, although the number of individuals seems relatively high, because of extent of occurrence and anthropogenic threatening factors like agricultural and mining activities *C.damboldtiana* is re-evaluated in category CR on the basis of IUCN Red List Categories and Criteria.

**KEYWORDS:** *Campanula damboldtiana*, Ankara, endemic, IUCN, conservation, Bern Convention

Poster presentation 20 05 37

### CREPIS PURPUREA, AN EXTINCTION-PRONE SPECIES FROM TURKEY

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*Crepis purpurea* (Wild.) M. Bieb. (Asteraceae) is endemic to the Crimean peninsula and has a minor variant in this area. This species has also been recorded with a brief description in the Flora of Turkey and the East Aegean Islands from Turkey. This study concerns the evaluation of morphological charac-

ters, current distribution, population status and the threats to the habitat of *C. purpurea* in Turkey with field and herbarium studies. An extended description of *C. purpurea* is presented here for the first time. The morphological characteristics of *C. purpurea* in Turkey are similar to those of its typical form in Crimea. *Crepis purpurea* is a critically endangered species with a single locality in Turkey. It is distributed in the Irano-Turanian phytogeographical region, including central Anatolian province in Turkey. Due to the anthropogenic pressures, fragmentation and erosion are present within the habitat of *C. purpurea*. Therefore it is evident that *C. purpurea* population is prone to extinction from the Turkish flora in the near future, unless *in situ* and *ex situ* conservation strategies are established as soon as possible.

**ACKNOWLEDGEMENTS:** This research was supported by the Scientific and Technological Research Council of Turkey (Tubitak, project no. 112T132).

**KEYWORDS:** *Crepis*, morphology, critically endangered, distribution pattern, Turkey

Poster presentation 21 05 24

### NEW DATA ON THE DISTRIBUTION OF *MARSILEA QUADRIFOLIA* AND *SONCHUS PALUSTRIS* IN BULGARIA

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*Marsilea quadrifolia* and *Sonchus palustris* are two species of Bulgarian flora with very few localities and are of national and international conservation importance. The aim of this study is to contribute some new data to the long-term studies of populations of these conservationally important species for providing actual data for their protection and management. Floristic studies and mapping of wetlands, fishponds, canals, and rice fields, carried out in the lowland parts of Bulgaria in 2014-2017. As a result from the studies two new localities (one for each species) of *Marsilea quadrifolia* (*Marsileaceae*) and *Sonchus palustris* (*Compositae*) were found in the Thracian Plain floristic region of Bulgaria. The plants of *Marsilea quadrifolia* were found to grow in a good condition on an area of about 3 square metres at the edge of a production rice field filled with water, situated between village Kostievo and town of Plovdiv at an altitude of about 180 m (N 42.16182° E 24.66838°). *Marsilea quadrifolia* has many known localities in other floristic regions: North-Eastern Bulgaria, Danubian Plain, Struma Valley (South), but most of them have not been confirmed during the last 60-80 years. The other known localities from Thracian Plain also remain unconfirmed at present. The only other one recently confirmed locality is in a drainage canal near Obnova village. Thirty five plants of *Sonchus palustris* were found to grow in a good condition on an area

of about 0.8 ha near the river Mechka at an altitude of about 240 m (N 41.97897° E 25.11764°), between the villages Leno-vo and Poroyna (Plovdiv district). Many of the plants were flowering and the tallest plant was 3.72 m high. *Sonchus palustris* is known from three floristic regions in the country – Black Sea coast, North-Eastern Bulgaria (lake Srebarna), and Danubian Plain, with several small populations. Most of the localities have not been confirmed during the last years. The new locality adds a new floristic region to the distribution of this species.

**KEYWORDS:** rare and endangered species, distribution, wetlands

Poster presentation 22 05 17

### INVESTIGATION OF THE POPULATION STATUS OF THE CRITICALLY ENDANGERED PLANT SPECIES *CENTAUREA WAGENITZIANA* (ASTERACEAE)

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*Centaurea wagenitziana* Bancheva & Kit Tan (Asteraceae) is one of the rarest plant species in the world. The species has a very limited distribution and is a tertiary relic. Several populations of the species have been known in the past from Bulgaria and Turkey (the region of Istanbul), but today it is found only in two localities situated on the territory of Southeast Bulgaria. It grows at the edge of shady meadows in xerophilous forests. The main threats to the species are: loss of the lowland oak forests in the past decades; various infrastructure projects in the area of the populations; overgrowing with bushes; low recruitment and migration potential of the species; specific habitat requirements; fires; massive infection by insects, etc. In order to protect the species from extinction a number of measures have been taken. As part of this study, an 8-year monitoring has been conducted to identify trends in population development, timely recording of threatening factors, and ongoing action to improve the current status. Thus, the number of the first population in 2018 amounts to about 250 individuals (a reduction of about 50% compared to 2010) and in the second one about 40 individuals were found (an increasing of about 25-30% compared to 2010). In 2017, in the implementation of the Action Plan, *in situ* activities at the first population were carried out to cutting bushes and branches. As a result of these activities, in 2018 twenty new seedlings in the control plots were registered. In addition, the individuals in shrub-cleaned areas are much more vibrant and in better condition than the control. The next step to be taken is to organize the relocation of all individuals from outside the protected site, so to be planted in suitable locations in the protected site.

**ACKNOWLEDGEMENTS:** Financial support by the National Fund "Scientific Research" of Bulgaria, project ДН-01/7/16.12.2016 'Flora of the Republic of Bulgaria, vol. 12: Biological Diversity in Asteraceae subfam. *Carduoideae* and *Cichorioideae*' and project №11233/ 10.08.2016 of Enterprise for the management of environmental protection activities, is gratefully acknowledged.

**KEYWORDS:** Asteraceae, *Centaurea wagenitziana*, conservation actions, endangered plants, Micro Reserves, population status

Poster presentation 23 05 07

### CRAMBE MARITIMA L. CONSERVATION IN ROMANIA

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*Crambe maritima* L. (seakale) belonging to Brassicaceae family is a perennial psammophyte and xeromezophyte species with deep root system, waxy leaves and scented white flowers that occurs on coastal habitats of the north-eastern Europe as well as on the south, west and north Black Sea coast. This plant grows on air-borne or soil-borne salty habitats, being a potential salt-tolerant crop with ornamental and economic potential. It is an edible plant species with young leaves similar to cabbage and young inflorescences may be consumed as broccoli. Also, young shoots buried-bred are edible for humans similar to Asparagus. Along the Romanian Black Sea coast, seakale is rarer compared to Bulgaria and it is endangered. Widespread in the past, nowadays it can be found only in few insular locations in counties Tulcea and Constanța, its habitats being lost and destroyed because of high degree of anthropization and dramatic reduction of wild beaches. Taking into account its conservative status and the diminishing of the populations, *in situ* and *ex situ* conservation strategies have to be implemented. The *in situ* protection measures involve the limited access of tourists in the growing area and *ex situ* preservation through the cultivation in botanical gardens and elaboration of optimized *in vitro* techniques. An *in vitro* multiplication method through somatic embryogenesis and direct morphogenesis can supply plants of *C. maritima* for repopulation.

**KEYWORDS:** *Crambe maritima* L., Black Sea coast, endangered, habitat destruction, *in situ*, *ex situ* conservation

Poster presentation 24 05 38

### CRITICALLY ENDANGERED, ENDANGERED AND VULNERABLE TAXA IN FLORA OF SPECIAL NATURE RESERVE ZASAVICA

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Nature reserve Zasavica is an peat swamp komplex formed by the rivers Drina and Sava in Atlantic faze of holocen and has total area of 1850 ha. Water system of Zasavica today gets its fresh water solely through underground streams of rever Drina. By the past 20 years of floristic reasearch in the Nature reserve Zasavica there was some 657 taxons registered. From 657 plant taxons recorded in the Nature reserve, we separated 45 taxons of the most important vascular flora. By the preliminary red list of flora Serbia and Montenegro in the flora of the reserve there are 27 taxons with the status of critically endangered (CR), endangered (EN), vulnerable (VU) and combined category of endergement : CR-VU (DD), EN-VU (DD), VU-NT (DD), VU-LC (DD). From total of 27 taxon plants, one or 3,7 % is in category of critically endangered (CR) [*Aldrovanda vesiculosa*], eight or 29,6 % is in the category of endangered (EN)[*Cardamine parviflora*, *Hippuris vulgaris*, *Hottonia palustris*, *Lindernia procumbens*, *Potamogeton acutifolius*, *Ranunculus lingua*, *Urtica kioviensis*, *Alisma graminea*], eight or 29,6 % in category vulnerable (VU) [*Dryopteris carthusiana*, *Leucojum aestivum*, *Stratiotes aloides*, *Thelypteris palustris*, *Tripolium pannonicum* subsp. *pannonicum*, *Batrachium aquatilis*, *Hypericum androsaetum*, *Ranunculus flammula*], one or 3,7 % in category CR-VU (DD) [*Schoenoplectus triqueter*], four or 14,8 % in category EN-VU (DD) [*Anacamptis palustris*, *Callitriche palustris*, *Potamogeton trichoides*, *Utricularia australis*], four or 14,8 % in category VU-NT (DD) [*Arum orientale*, *Erysimum cheiranthoides* subsp. *cheiranthoides*, *Hesperis sylvestris* subsp. *sylvestris*, *Thymelaea passerina*], one or 3,7 % in category VU-LC (DD) [*Zannichellia palustris*]. Most important taxon in the reserve is *Aldrovanda vesiculosa* for witch SNR Zasavica is the only habbitat in Serbia. These results clearly indicates national and international importance of this nature area and because of that Zasavica is one of the IPA areas in Serbia and future Natura 2000 and Emerald areas.

**KEYWORDS:** Zasavica, critically endangered, endangered, vulnerable taxa, flora

Poster presentation 25 05 10

### GERMINATION OF GYMNADENIA CONOPSEA MATURE SEEDS (L.) R.BR. BY ASIMBIOTIC CULTURE IN VITRO

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Although family *Orchidaceae* contributes 10% to the total number of flowering plants, approximately quarter of established species are endangered, as a consequence of habitat loss, plant smuggling, pervasive impact of global climate change as well as overexploitation for horticultural purposes. In this manner, *in vitro* propagation techniques play an irreplaceable role in their conservation. Efficiency of this technique depends of many factors which must be adjusted to each species. The aim of this study was to establish asymbiotic germination protocol for the purpose of *ex situ* conservation of endangered orchid species *Gymnadenia conopsea* (L.) R.Br. Influence of two basal media - Knudson C (KC) and Malmgren (MM) – and organic additives - coconut water (CW) pineapple juice (PJ), peptone (PE), L-glutamine with folic acid (A) and casein hydrolysate (CA) (for KC medium) – were tested. All tested media consisted of 2% sucrose, 7% agar and 1% activated charcoal, while the pH for all media was adjusted to 5,8±0,02 before autoclaving at 121 °C for 20 min. Influence of illumination was tested by placing the Petri dishes with seeds under two different light conditions: 16/8 light/dark and 0/24 light/dark (continuous darkness). The results showed that MM medium was more effective than KC medium for germination, protocorm, shoot and rhizoid formation. All organic supplements accelerated protocorm formation, while the highest germination efficiency was observed on MM-CW medium. Rapid germination, growth, and development occurred in continual darkness, contrary to 16/8 light/dark which promoted only swelling of the embryo.

**KEYWORDS:** *ex situ*, conservation, orchids, terrestrial, germination

Poster presentation 26 05 30

### TESTING THE SEED GERMINATION OF OXYTROPIS PILOSA (L.) DC. 1802 (FABACEAE)

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*Oxytropis pilosa* (L.) DC. is a species which is widely distributed in Central and East Europe, with populations found in the Balkans. It is one of the rarest species with the Pontic distribution type. Due to intensive habitat loss, it is classified as an endangered species. As most species of the Fabaceae family, *O. pilosa* expresses a physical type of seed dormancy. In order to break this dormancy, seeds must be exposed to a specific combination of ecological factors, which induce changes in the seeds and trigger their germination. The objective of this research was to show under which circumstances seeds of this species successfully germinate. In this paper, we tested the germination of the species' seeds after applying three treatments to break their dormancy. These treatments were: scarification by boiling water, scarification by sulphuric acid and mechanical scarification. The effects of these treatments were compared to germination success in the control group (without treatment). Also, in order to test the seed germination, we formed three experimental groups, on which top-of-paper method of seed germination was performed. Two of these experimental groups were made of seeds which weren't mechanically scarified; One group was kept exposed to daylight, while the other was kept in the dark. The third experimental group was made of seeds, which were mechanically scarified and kept in daylight. Seeds, which were mechanically scarified showed the highest level of germination, what could serve as a proof of physical dormancy type of this species' seeds. Seeds in the control group also successfully germinated, despite their dormancy. This can imply that this species' seeds would not need any specific ecological circumstances to germinate in situ. Seeds, which were treated with boiling water and sulphuric acid showed very low germination rates, which could be a result of embryo damage. We conclude that the application of mechanical scarification could be an effective method to induce seed germination in future conservational research of this species. The endangered conservation status of *O. pilosa* may not be the result of seed dormancy, but a consequence of habitat destruction.

**KEYWORDS:** endangered species, germination, Leguminosae, seed dormancy, seed scarification

Poster presentation 27 05 12

### VALUES FOR NATURE CONSERVATION: LOCALS' VIEWS ON THE IMPACTS OF TRADITIONAL PIG HERDING ON FLOODPLAIN FORESTS ALONG THE SAVA RIVER

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The Bosut forest, with its exceptional biodiversity depending partly on traditional livestock farming, is a model site of European importance. It could show how the new nature protection paradigm based on cooperation and knowledge co-production with local inhabitants can be more effective than the previous „fortress” model. In 2017, 14 semi-structured interviews were conducted with pig herders and local foresters to reconstruct the changes in the grazing regime and the ecosystem services gained from traditional pig keeping in the Bosut forest, and 8 interviews were made with pig herders to document their knowledge of forest and marsh plants and habitats. Locals knew more than 150 plant species, and had a deep understanding of the pigs' foraging preferences (e.g., by species and by season). We learnt that locals had rights to graze pigs in the forests since time immemorial. Main regulatory rules of forest grazing have not changed in the last decades. Many locals prefer forest-reared piglets during family festivities because it is, as they argue, more healthy and tasty. They emphasized that forest pigs can utilize the acorn production and also the plants of the herb layer and convert it to meat for human consumption. Most locals argued that pig grazing is beneficial to the forest. For example, pigs eat up larvae of pests thus decreasing their impact on trees, especially oaks. Pigs loosen the soil thus help infiltration of the water, so the forest floor is less dry in summer. Their opinion is that pig grazing is a natural phenomenon, so it is good for Nature. Local foresters and herders perceived no negative impact of grazing on the natural values in marshes or in forests. Herders argued that pigs keep the marshes free of bushes and prevent their closure by *Phragmites*, *Carex* spp. and *Glyceria maxima*. The practices and knowledge of pig grazing in the Bosut forest is an intangible cultural heritage of European importance. This is one of the very last places in Europe where the previously wide-spread forest pig keeping survived and can be protected and studied by botanists and conservationists. The related traditional ecological and vegetational knowledge is a valuable knowledge bank (analogue to a gene bank).

**KEYWORDS:** floodplain forests, pig grazing, local culture, ecosystem services, conservation

Poster presentation 28 05 14

### TRADITIONAL PIG HERDING IN THE FLOODPLAIN OF THE SAVA RIVER - AN ECOLOGICAL PERSPECTIVE OF FOREST AND WETLAND GRAZING IN BOSUT FOREST, SERBIA

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Traditional pig herding in hardwood floodplain forests of Europe has a long history. Feeding pigs on acorn, fruits, forest grasses, worms and roots was institutionalized in the practice of 'pannage' in the Middle Ages. However, we have little knowledge about how these disturbances drive the development of the structure and dynamics of forest and wetland communities. Therefore, Bosut Forest Area represents a unique socio-ecological system where pannage and pig grazing in forests and wetlands is still alive. The aim of this paper is to present the results of our research on the impacts of pig activity. In the autumn of 2017 and spring of 2018 vegetation surveys and interviews with pig herders were performed to document the effect of pig herding on the herbaceous and shrub layer attributes of hardwood floodplain forests and wetlands. Hydrological regime, available light and pig grazing have been forming and developing the structure and dynamics of forest and wetland communities of Bosut Forest Region. Forest and marshland structures are very sensitive to the changes of grazing pressure and flooding regime. High disturbance intensity by pigs is limited to small areas in the forest, where pigs create and maintain unique microhabitats (e.g. muddy ponds, puddles, wetlands or small openings near the 'salaš' places). Some of these microhabitats provide habitat for valuable plant species such as *Marsilea quadrifolia*, *Hottonia palustris*, *Ludwigia palustris*, *Lindernia procumbens*, *Elatine triandra* and *E. alsinastrum*. Grazing, trampling and rooting activity of pigs can contribute to the development of a diverse pattern of shrub layer structure in the forest and preventing shrub encroachment in marshes. The abandoned wetlands are threatened by shrub encroachment, mostly with *Amorpha fruticosa*, *Fraxinus angustifolia ssp. oxycarpa*, *Crataegus laevigata*, *C. monogyna*, *Salix cinerea*, *Pyrus ppyraster*, *Malus sylvestris*, and sporadically in other species. Keeping alive and enhancing traditional farming activity in the Bosut area would be a cost-effective way of habitat maintenance for nature conservation and would contribute to the sustaining of an old skill and the related traditional ecological knowledge, both of which are of European importance.

**KEYWORDS:** Floodplain, oak forests, wetland, pig grazing and rooting, ecological impact

Poster presentation 29 05 25

### PRELIMINARY CHECK-LIST OF INVASIVE ALIEN PLANT SPECIES (IAS) IN REPUBLIC OF MACEDONIA

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In this paper a preliminary check-list of invasive alien plant species (IAS) in Republic of Macedonia is presented. Alien species are considered to be »species introduced and growing outside of their natural distribution«. The problem of IAS has become one of the most important issues in conservation biology during the last decades. Invasive species are nowadays recognized as the second most important cause of biodiversity loss, just after direct habitat destruction. The preliminary check-list has been created on the basis of the literature and of field observations. The list consists of 54 taxa, with family, life-form and origin were attributed to each IAS given on the check-list. The life-forms were interpreted according to Raunkier's system denoted with the following abbreviations: P – Phanerophytes, Ch – Chamaephytes, H – Hemicryptophytes, T – Therophytes, G – Geophytes, Hy – Hydrophytes. Geographic origins of IAS were attributed only in general terms (at the continental level except for Mediterranean species) and were assigned with the following abbreviations: Am – the Americas (North and South), As – Asia, Af – Africa, EA – Eurasia, M – Mediterranean. Data on origin were taken mostly from the available literature. Basic statistical analyses were carried out to show the relative abundance of IAS according to the family affiliation, life-form and geographic origin. Taxa that originate from two different continents were considered as representatives for each of them in the origin analysis. Out of 26 families the majority (22 families) belong to the dicotyledons. The most numerous family is Asteraceae, and genera with the highest number of IAS are *Erigeron*, *Amaranthus* and *Solanum*. Life-form analysis showed the predominance of therophytes but also presence of hemicryptophytes, phanerophytes and geophytes. In the origin analysis, IAS from the Americas predominated, followed by those from Asia and Africa.

**KEYWORDS:** invasive alien plant species (plant IAS), alien flora, introduced flora, Republic of Macedonia

Poster presentation 30 05 03

### INVASIVE PLANT SPECIES IN SERBIA AND NEIGHBORING COUNTRIES

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The aim of this paper is to establish the presence of invasive plant species in countries surrounding Serbia (Hungary, Romania, Bulgaria, Macedonia, Montenegro, Bosnia and Herzegovina, Slovenia and Croatia) and assess the possibility of their intentional or unintentional introduction or migration to Serbia. In order to control the transport and/or prohibit the invasive plant species introduction from neighboring countries to Serbia, first of all it is important to establish which invasive species from these countries can be successfully inhabited, reproduced and dispersed in Serbia. If their movement routes and pathways are determined, it is possible to react in a time and stop their spreading. The data on the presence of invasive species in Serbia and eight neighboring countries were collected from the available literature for the period from 2008 to 2017. The list of common invasive species for all of these countries was established and compared with the DAISE (Delivering Alien Invasive Species Inventories for Europe) list "100 of the Worst" and the list of Invasive Alien Species of Union concern. The total number of invasive plant species was 188, in all 9 countries. The species *Duchesnea indica*, *Chamomilla suaveolens*, *Conyza bonariensis* and *Paspalum paspalodes* are invasive in many neighboring countries, but in Serbia these species are not invasive. The proposal is to add these 4 species on the list of potentially invasive species of Serbia. It is interesting that species *Sisyrinchium montanum*, *Sisyrinchium bermudiana* and *Laburnum anagyroides* are invasive in Bulgaria and Romania, but they are strictly protected in Serbia.

**KEYWORDS:** invasive plant species, neighboring countries, Serbia

Poster presentation 31 05 39

### THE URBAN FLORA OF IMOTSKI

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Recently, the flora of the cities of many European countries has been explored to identify new plant species that have a positive or negative impact on the existing flora. In this paper, the urban flora of the town of Imotski was studied with the aim to provide a list of autochthonous and allochthonous

plant species, taxonomic analysis of flora and habitat types. The identification of collected specimens has been done with the help of standard floras. The urban flora of Imotski contains 321 plant species of vascular plants distributed on 9 anthropogenic ruderal habitats. The most dominant families in the investigated flora are Asteraceae (48.5%) and Poaceae (43.0%). The dominant life-form categories of flora in the area of Imotski are terophytes (37.5%). The analysis of floral elements shows that wide spread plants (26.6%), Mediterranean (23.0%), cultivated and adventive plants (22.4%) are most represented. Dominant plant species on ruderal habitats are *Ailanthus altissima*, *Polygonum aviculare*, *Cynodon dactylon*, *Portula oleracea*, *Setaria viridis*, *Chenopodium album* and *Conyza canadensis*. According to the total number of species in flora, 11% of the species belongs to the allochthonous adventive plants (neophytes), which are constantly spreading into the composition of natural and anthropogenic habitats. Most of the neophytes originate from the American continent (22 plant species or 63.0%). One of the most invasive species in the flora of Imotski as well as in the flora of Croatia is *Ailanthus altissima* which spreads to almost all habitats of the urban flora of Imotski. In the investigated flora of Imotski, two endemic species belonging to the Illyrian Adriatic endemic plants and four endangered plant species were found. The influence of man and further urbanization decreases the natural habitats and increases the proportion of ruderal habitats in the flora of Imotski with allochthonous flora.

**KEYWORDS:** flora, Imotski, allochthonous species, neophytes, invasive species

Poster presentation 32 05 23

### ENDANGERED SANDY COAST IN VELIKA PLAŽA (MONTENEGRO)

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Velika plaža is the large sandy coast in the Eastern Adriatic, occupying the southernmost part of Montenegrin coast, near the Albanian border. In the last few years we conducted several studies along the whole length of the coast, including all important habitat types along the seaside-inland gradient: annual vegetation on drift lines (1210), embryonic shifting dunes (2110), shifting dunes along the shoreline with *Ammophila arenaria* (2120), humid dune slacks (2190), wooded dunes with *Pinus pinea* and/or *Pinus pinaster* (2270). In the years 2016-2017 we recorded plant species composition and cover, beach litter composition and cover in 2x2 m plots, and tried to link the results with the estimates of anthropogenic activities, all of it with the emphasis on the differences between individual habitat types. We found that i) trampling by humans and

grazers significantly alters the species composition and the installation of simple fences helps to restore the richer communities, ii) number of alien taxa is not high (*Erigeron annuus*, *E. canadensis*, *E. sumatrensis*, *Oenothera* spp., *Sporobolus indicus*, *Xanthium orientale* ssp. *italicum*), but they are widespread and the cover may be high, except in the humid dune slacks, where they are rare; iii) alien species change some functional and phylogenetic diversity indices in the stands where they occur; iv) beach litter is widespread across all habitat types in at least small quantities, but frequently the cover and diversity of litter is very high; the only exception are the humid dune slacks with very low litter quantities; v) source of litter is different in different habitat types. Every year there is a visible “development” of tourist facilities - installation of new beach cottages/huts, construction of new roads to the beach area. With all of the mentioned above we can conclude that there are several aspects of endangerment on Velika plaža that need to be taken into account when planning future touristic development, especially considering it's the northernmost larger preserved sandy beach on the Eastern Adriatic coast.

**KEYWORDS:** Velika plaža, sandy beach, alien species, beach litter, human impact

Poster presentation 33 05 40

### PLANT SPECIES SIGNIFICANT FOR THE BIODIVERSITY CONSERVATION IN THE CANAL NETWORK OF BANAT (VOJVODINA PROVINCE, SERBIA)

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Due to its exceptional floral wealth and a large number of endemic taxa, the Balkan Peninsula is one of the world's major centers of biodiversity. The aim of the study is to point out the presence of rare and endangered plant species of national and international significance in the canal network of Banat (Vojvodina Province, Serbia). Due to the significant negative influence on the biodiversity, the presence of invasive species for the territory of Vojvodina/Serbia and Europe, as well as invasive species of global importance in the canal network have been pointed out. During the floristic studies, a total of 178 taxa were found, of which 166 species are on one of the protected and/or invasive species lists for the territory of Serbia and/or Europe. Eighteen species are protected in Serbia, while 111 are on the IUCN list of endangered species of Europe (most with Least Concern status). Thirteen species are invasive for the area of Vojvodina (IASV list), 115 are invasive in Europe (DAISIE list), and 28 species are invasive species of global importance (GISD list). The most widespread endangered species in the canal network of Banat are: *Iris pseudoacorus* L., *Potamogeton nodosus* Poir. and *Trapa natans* L., and from invasive species of Vojvodina and Europe: *Amorpha*

*fruticosa* L., *Calystegia sepium* (L.) R. Br., *Glyceria maxima* (Hartm.) Holm., *Lemna minor* L., *Lythrum virgatum* L., *Rumex hydrolapathum* Huds., *Salix alba* L. and *Typha angustifolia* L. A significant number of species from the endangered species list of Europe is not endangered and does not enjoy special protection in Serbia, so it is necessary to harmonize the mechanisms of protection at the national levels with European and global mechanisms of protection. In addition, a large number of species endangered in Serbia and/or Europe are also on the list of invasive species of Europe, and it is necessary to harmonize the existing lists of endangered and invasive species of Europe. Bearing in mind the richness of the flora of the Balkan, Serbia's responsibility for preserving biodiversity is significant.

**KEYWORDS:** protected and endangered plant species, invasive plant species, Serbia, Europe, biodiversity, canal network

Poster presentation 34 05 26

### FIRST REPORT OF ESCAPED OCCURRENCES OF THREE ALIEN ORNAMENTAL TREE SPECIES IN BULGARIA

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Ornamental plants are a major source of escaped, and consequently naturalized, occurrences of many alien species in most regions of the world. In Bulgaria, numerous tree species were introduced as ornamental plants in the late XIX and first half of the XX century and some of them became established and even invasive, for example, *Acer negundo*, *Ailanthus altissima*, *Broussonetia papyrifera*, *Fraxinus americana*, *F. pennsylvanica*, *Gleditsia triacanthos*, *Robinia pseudoacacia*. The objective of this study is to report for the first time naturalized occurrences of three alien phanerophyte species in Bulgaria – *Catalpa bignonioides*, *C. ovata* (Bignoniaceae) and *Paulownia tomentosa* (Paulowniaceae). These species have been planted as ornamental trees in many parts of the country, mainly in larger towns. The distribution data has been collected by the authors during floristic studies throughout the country over the past decade. So far the three species have been recorded mainly in managed environments – urban areas. However, bearing in mind the readily available saplings from larger garden centers and the relatively mild winters in most parts of Bulgaria during the past decade, it is projected that the naturalization of the three species will continue. Moreover, in some of the naturalized occurrences, e.g. of *P. tomentosa* in the Black Sea Coast (Northern) and Northeast Bulgaria floristic

regions, the naturalized plants are already at a reproductive age and form small self-established populations.

**KEYWORDS:** alien species, Bignoniaceae, *Catalpa*, *Paulownia*, Paulowniaceae, ornamental trees

Poster presentation 35 05 20

### POTENTIAL DISTRIBUTION OF GOLDEN CROWNBEARD (*VERBESINA ENCELIOIDES*) IN ROMANIA

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Romania is one of the countries in South-eastern Europe, where special interest is given to the alien species with potentially invasive effects. Preventing the introduction of these species is preferable to intervention or post-invasion control. The challenge is to identify the species beforehand in order to avoid its dispersal, as well as areas exposed to such risks. Since the areas prone to be invaded by alien species is dynamic, the development of models that predict the expansion and identify migration corridors and risk areas are extremely useful in developing policies to control and manage invasive species. *Verbesina encelioides*, a species of North American origin, has been recently reported in Romania's flora. The naturalization and the rapid expansion of this species determined us to research its invasive potential by modelling the potential species distribution at a national level using MaxEnt algorithm, bioclimatic and landuse variables. The result may be useful in explaining if climate change may affect its expansion and also may help identifying hotspot areas and thus control the spread of this species.

**KEYWORDS:** *Verbesina encelioides*, invasive plant, potential distribution, maxent

Poster presentation 36 05 41

### NEW SITES OF ALLOCHTHONOUS SPECIES *CYPERUS ODORATUS* L. (FAM. CYPERACEAE) IN THE MIDDLE FLOW OF THE DRINA RIVER WATERSHED

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*Cyperus odoratus* L. is a pantropic species, naturalized in many parts of southern Europe, and is often mistaken for species *Cyperus strigosus* L. and other similar species. The data collected so far indicate that this allochthon species was found in Serbia on the Danube section from Kovin to Mihajlovac. During the research of the riparian areas along the Drina River and its tributaries in 2016. and 2017., the species *Cyperus odorata* L. was found on five locations: on the Radaljska river bank (September 30, 2017.), on the bank of the Ljubovidja river (September 22, 2016.), along the bank of Velika Reka near the mouth in to the Drina river (September 21, 2016.), along the Gračanička river bank, 200 m before the entrance to Drina (September 23, 2016.) and on the bank of Drina river at in Parašnica Crna bara (August 8, 2017.). A small number of specimens was found in all the locations (1-3). The following dominant plant species that were recorded in the proximity of the fish species mentioned above: *Juncus articulatus*, *Juncus effusa*, *Alisma plantago-aquatica*, *Bidens* sp., *Veronica anagallis-aquatica*, *Eupatorium cannabinum*. The finding of this species along the Drina River complements the knowledge of the distribution of this allochthonous species in Serbia and indicates its spread south of the Sava and the Danube.

**KEYWORDS:** *Cyperus odoratus* L., Drina River watershed, allochthonous species

### THE INFLUENCE OF PHYSICAL-CHEMICAL WATER PROPERTIES TO THE DEVELOPMENT OF INVASIVE COMMUNITIES *ELODEETUM CANADENSIS* AND *CERATOPHYLLO DEMERSI-VALLISNERIETUM SPIRALIS* IN HYDROMELIORATIVE FACILITIES IN BAČKA (SERBIA)

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The occurrence of allochthonous species in aquatic ecosystems is not always a cause for concern. However, a large number of these species can become invasive. Invasive macrophytes negatively change ecosystem properties and complex interactions in them. They are identified as the main cause of loss of biodiversity. Invasive species also affect the functionality of watercourses because the production of huge biomass of these species can: prevent optimal flow in the watercourses, limit the use of water for recreational purposes, disturb the light regime, prevent water mixing and cause changes in many physical-chemical water properties. The Water Framework Directive (EU 2000) explicitly states that their presence must be taken into account when assessing the ecological status of surface waters. In many European countries, the occurrence of invasive species is well documented, but for the vast majority of these species, data on their quantitative representation, as well as the degree of invasion, are missing. Phytocoenological studies provide valuable data about the invasiveness degree, which have an adequate importance in assessing the effects on the diversity of autochthonous vegetation. The aim of this study is to define the basic physical-chemical water properties influencing to the development of two invasive phytocenoses (*Elodeetum canadensis* and *Ceratophyllo demersi-Vallisnerietum spiralis*), an analysis of their floristic structure and distribution in the canal network in Bačka. Physical-chemical water properties were determined according to standard methods. The following properties were monitored: temperature, pH value, total alkalinity, chemical (COD-MnO<sub>4</sub>) and biological (BOD<sub>5</sub>) oxygen demand, and nutrient content (NH<sub>4</sub><sup>+</sup>, NO<sub>3</sub><sup>-</sup>, NO<sub>2</sub><sup>-</sup>, PO<sub>4</sub><sup>3-</sup> and total P). The properties determining the differentiation of the phytocenosis *Elodeetum canadensis* and *Ceratophyllo demersi-Vallisnerietum spiralis* are pH, BOD<sub>5</sub> and NO<sub>2</sub><sup>-</sup>. The *Ceratophyllo demersi-Vallisnerietum spiralis* stands, compared to the *Elodeetum canadensis* stands, prefer water with higher values of pH and BPK<sub>5</sub> and lower values of NO<sub>2</sub><sup>-</sup>. Phytocenosis *Elodeetum canadensis* ascertained only in the canal "Kosančić-Mali Stapar" and it has no tendency to spread, for now. Ass. *Ceratophyllo demersi-Vallisnerietum spiralis* stands are found on several sites and have a tendency to spread, so the monitoring of these stands is necessary.

**KEYWORDS:** invasive species, properties of water, eutrophication, macrophytes

### INVASIVE PLANT CAN BE USEFUL – EFFECTIVE AWARENESS RAISING THROUGH THE PROJECT APPLAUSE

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Invasive alien plant species (IAPS) are one of the main threats to biodiversity worldwide. Their presence in urban and agricultural areas can also cause significant economic damage. Raising awareness of the harmfulness of invasive species among various interest groups has proven to be a challenging task. There are known examples of good practice from various countries, on raising awareness of IAPS and educating the population on their eradication. In most cases, the removed plants are composted or incinerated, which has been the prevalent practice in City of Ljubljana as well. With the pilot project for semi-industrial production of paper from selected IAPS, we have already proven that these plants can be utilised usefully. The project »APPLAUSE – from harmful to useful with citizen-led activities« is the next step which addresses unresolved issues about invasive alien plant species in terms of the zero-waste approach and circular economy. The proposed IAPS management system is based on education and cooperation with the citizens of Ljubljana, and three principles of operation: 1. Do it yourself, 2. Process with us and 3. Bring to the collection centre. We will develop a more environmentally friendly method for pretreating the pulp for paper production – processing with enzymes. Additionally, the cooking liquor (waste from delignification process) will be processed into a raw material for industrial production. We will also develop solutions for processing wood waste into useful products. We will evaluate the suitability of selected IAPS for human consumption, for production of dyes and for home-made anti-pest agents. We will develop a business model that can be applied internationally for creation of new green jobs and inclusion of long-term jobseekers and people with disabilities. By introducing handicraft workshops in the reuse centre, we are preserving traditional crafts, cultural heritage and knowledge. In addition to field surveys of the distribution of IAPS in the City of Ljubljana, we will use data from the new generation of European Earth observation satellites Sentinel-2, and other information and communications technologies.

**KEYWORDS:** invasive plants, circular economy, applause, zero-waste, awareness raising

### RAGWEED (*AMBROSIA ARTEMISIIFOLIA*) CONTROL SYSTEM IN URBAN AREAS OF THE CITY OF NOVI SAD

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Ragweed (*Ambrosia artemisiifolia*), is an invasive, allergenic weed plant widely spread throughout the territory of the Republic of Serbia. Despite its negative impact on human health, its propagules often find suitable surfaces for establishing new colonies, even in areas where still preserved, though fragmented, natural plant stands thrive. The City of Novi Sad has been expanding to its surrounding agricultural zones through the process of urbanization. Such zones are quite often characterized by ruderal habitats, from which ragweed can spread back into urban areas. Based on a decade-long experience ragweed monitoring and control in the Novi Sad in urban areas, a ragweed spread control system has been established and applied. This control system aims to reduce the concentration of air borne pollen and generally remove/reduce the number of ragweed plants in urban public areas. The only certain way to monitor the ragweed population and control its spontaneous spread is a consistent implementing of a control system - successive mechanical plant removal in regular and strictly defined time intervals before the flowering begins. The ragweed control program enables several mowings of such surfaces, from June to October. Conducting multi-annual monitoring has led to the categorization of the ragweed-present areas, thus such established monitoring model facilitated the triage of the ragweed-present areas and the determination of priorities in further ragweed monitoring and control.

**KEYWORDS:** ragweed, monitoring, control system

### COMMON RAGWEED – INVASIVE WEED ON THE TERRITORY OF NOVI SAD

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In recent decades problem of invasive weed species is increasing throughout the world, as well as in our country. Invasive weed species are considered to be those species that are intentionally or accidentally transferred from their natural habitats into new ecosystems, where in competition process they suppress indigenous (native) species conquering available surfaces. During eight years lasting studies on the territory of the city of Novi Sad, terrain studies of *Ambrosia artemisiifolia* L. distribution as well as mapping were performed on regulated and disordered green areas and on arable areas. For mapping of ragweed distribution, partially modified method of Braun-Blanquet was used and data processing was performed by specially designed program *Ambrosia Spot Marker*. Monitoring of this species was also done because of the occurrence of retrovegetation after mowing. Every year we made the monitoring of this invasive species on about 475 ha of ruderal and suburbs areas on the territory of Novi Sad. It revealed the presence of *A. artemisiifolia* on over 200 locations in 13 city zones (Petrovaradin, Begeč, Budisava, Veternik, Kač, Kisač, Kovilj, Rumenka, Stepanovićevo, Futog, Čenej, Šangaj i Pejićevi salaši). After visiting all the suburbs recommendations for mechanical or chemical control of ragweed were forward. Petrovaradin, Šangaj and Begeč were areas with the highest number of established individuals per m<sup>2</sup>, as well as bank along the Danube river. These locations are extremely favorable for the spread of ragweed, because they are exposed to high daily temperatures. In other suburban areas ragweed established in landfills, access roads to the village, children's playgrounds, sport fields, parks and large public spaces. Common ragweed is determined at all ruderal habitats in the suburbs near Novi Sad, which presents a problem for human health and children due to pollen plants. In the last 8 years we have been monitoring the weed species on a regular basis and made recommendations for its mechanical and chemical control.

**KEYWORDS:** *Ambrosia artemisiifolia*, Novi Sad, ruderal areas, allergy weed, invasive weed

Poster presentation 41 05 44

**POTENTIAL RISKS CAUSED BY MARINAS  
TOWARDS LAND-SEA INTERACTIONS AND  
THE NATURE IN TURKEY**

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Nowadays, global warming and environmental awareness have been of great importance as a result of considerable environmental pollution, depletion of natural resources and unconscious management. The problem of global warming remains serious, although many authorities have taken action and lots of sanctions have been made in order to protect environment and decrease pollution. Tourism which is a driving factor for most of the countries' economies is very critical element for the environment as well. Especially sea tourism should be intensively concerned in order to control potential hazards towards land-sea interactions. Day by day, the number of marinas are increasing and the risk is getting high. In this study, a literature review was conducted to see global effects of marinas towards land-sea interactions. In addition, interviews were carried out with different professionals either working in marinas or working for sea tourism in Turkey. The main goal of the interviews was to understand the current situation and highlight the potential risks in land-sea interaction caused by marinas. In conclusion, it has been seen that most of the marinas which are mainly constructed in untouched nature places, constitute a considerable risk for the land, the forestry, plant species and the sea. While some authorities take preventive measurements, some others still do not. This brings about a big danger for the environment. One of the other problem is the leakage of substances from motorboats. The leakage causes not only pollution of the sea but also disturbance of ecological balance and botany invasion. Many plant species are in danger of disappearing due to uncontrolled marina activities. While the number of marinas are increasing, the authorities should take necessary actions and give more importance to the preservation of nature.

**KEYWORDS:** land-sea interactions, environmental awareness, risks caused by marinas, sea tourism, botany invasion

**Session 6.**  
*Phytochemistry and Plant Resources*



Balkan  
Botanical  
Congress

## PHYTOCHEMISTRY - BOTANIST'S POINT OF VIEW

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Plant chemistry or phytochemistry would be the appropriate names for the field of knowledge that we want to describe and clarify here. However, to the chemist, plant chemistry may have quite a different meaning than to the botanist. So, the term phytochemistry solves any possible confusion. Phytochemistry is a sub-field of Botany or Chemistry, represents the bridge which connects chemistry and botany. The subject of phytochemistry, or plant chemistry is between natural product organic chemistry and plant biochemistry and is closely related to both. As the subject of phytochemistry, various metabolites, which plants synthesize and accumulate, and which have special biological functions important for the adaptation and survival of plants, and which in many cases are under strict genetic control, it is clear that they are also of particular interest to botanists. Given that phytochemistry is deals with the chemical structures of these substances, their biosynthesis, turnover and metabolism, their natural distribution and their biological function, botanists, using phytochemical data, it can make important conclusions related to specialized fields of botany, morphology, physiology, ecology, systematics and phylogeny and the evolution of plants. On the other hand, men whose training is largely in chemistry, sometimes err in assuming that chemistry is a magical touchstone which will quickly explain everything and they consider themselves to be taxonomists (chemotaxonomists). Yes, they maybe resolve the chemical structure of the new compounds from plants, but they role in point of view in morphology, ecology, phylogeny and taxonomy must be a field of botanists. This leads to a slightly different point of view that, botanists (keeping in mind the specialized metabolites and their metabolic pathways, discovered by chemists) are those who, in light of the complex relationships between the plant organism and its environment - adaptation and evolution, can give an overview of whether these chemical metabolites are a valid tool for e.g. solving the status of problematic plant taxa, or what is their functional role.

**KEYWORDS:** phytochemistry, botany, point of view

## NEW APPROACH FOR PLANTS SUSTAINABLE EXPLOITATION - HEALTH TRACKS AND FOREST BATHING

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Although the beneficial effects of many medicinal and non-medicinal plants are well known, the rise of consumers' attitude for healthy lifestyle demands new approaches for plants utilization. The concept of forest bathing (Shinrin-yoku) has been recognized in Japan since 1982 and so far more than 60 forest therapeutic centers have been housed in the world. Also, all over the world many health tracks are mapped. However, the expansion of this concept slows down the complexity of the quantification of therapeutic effects of different ecosystems. Recent studies have revealed the acute psychological and physiological effects of benefits of green environments and most European health resorts established for rehabilitation and optimization of health conditions are located in natural environments. It is confirmed that natural environments prevent occurrence of pulmonary and cardiovascular diseases, hypertension, obesity, cancer, insomnia and anxiety, depression, burnout syndrome, as well as the concept of stress-related lifestyle. The trend of healthy living and returning to nature makes the expansion of rural and wellness tourism, one of the main modern developmental strategies worldwide. Many tourists are interested in visiting protected nature areas with air spas and anti-stress gardens. Regarding to vast plant biodiversity and numerous medicinal plants, as well as a large number of protected areas and national parks, Serbia has enormous potential for exploitation of the medicinal effects of stay or walk in the nature. However, the main problem is lack of proper education about the utilization of plants in tourism, as well as the absence of the appropriate marketing strategy. After overcoming these facts, establishment of health tracks and forest baths all over Serbia could notably increase the tourism profile and maximize the utilization of the previously unrecognized potential of plants for the tourism development.

**ACKNOWLEDGEMENTS:** Interreg-IPA CBC HUS-RB/1602/31/0075

**KEYWORDS:** green environments, sustainable exploitation, tourism

## ESSENTIAL OIL AND PODOPHYLLOTOXIN IN JUNIPERUS SABINA, J. EXCELSA AND J. VIRGINIANA IN BULGARIA

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The essential oil (EO) of juniper species has wide applications in various products. Podophyllotoxin is a toxin currently extracted from the Himalayan mayapple (*Podophyllum hexandrum* Royle), and is used as a precursor to the commercially available anti-cancer drugs etoposide and teniposide. Some junipers may also contain podophyllotoxin. The EO profile and podophyllotoxin concentration are function of juniper species, sex (most junipers are dioecious), but may also depend on subspecies/chemotype, the environment, the plant part from which it is extracted (leaves, galbuli, or wood), and the extraction procedure. The objective of the study was to assess variability in EO profile and podophyllotoxin concentration of three junipers that have limited distribution in the Bulgarian flora; *J. sabina* L., *J. excelsa* M. Bieb., and *J. virginiana* L. (introduced species, used in landscaping). The EO content in *J. excelsa* varied from 0.7 to 1.9%, whereas the EO content in *J. sabina* was 1.3-2.1%, and the EO content of *J. virginiana* was 0.9%. The major EO constituents of *J. excelsa* (in order of decreasing concentrations) were  $\alpha$ -cedrol,  $\alpha$ -limonene, and  $\alpha$ -pinene, the ones of *J. sabina* EO were sabinene, terpinene-4-ol, and elemol. The EO constituents of *J. virginiana* included  $\alpha$ -cedrol,  $\alpha$ -limonene, safrol, elemol and methyl eugenol. Podophyllotoxin was found in the leaves of *J. sabina* and *J. virginiana*, but not in the *J. excelsa* accessions. Overall, the essential oil profile of the three junipers was quite different and may satisfy the essential oil industry utilizing juniper leaf essential oil. However, *J. sabina* and *J. excelsa* are protected species in Bulgaria, and therefore, their natural populations may not be utilized for commercial production of EO of podophyllotoxin.

**KEYWORDS:** junipers, volatile oil, oil profile, podophyllotoxin

## SURFACE FLAVONOIDS AS CHEMOTAXONOMIC MARKERS IN DIFFERENTIATION OF SATUREJA MONTANA AND S. SUBSPICATA

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The genus *Satureja*, savory (Lamiaceae), is distributed in the Mediterranean, tropical Africa, Asia, and the Americas. The exact number of species is still uncertain, due to the considerable taxonomic confusion associated with the generic limits of the so-called *Satureja* complex. *S. subspicata*, now having the taxonomic rank of a species, was previously treated as a subspecies or variety of *S. montana*. In the late 1980's, first studies on the taxonomic importance of flavonoids in Nepe-toideae have appeared, showing genus-specific surface flavonoid profiles, but not many studies have focused on lower taxonomic levels. The literature survey has shown surface flavonoids have only been studied from one individual of *S. montana*. In order to validate the usability of surface flavonoids as chemotaxonomic markers in two phylogenetically very close species, *Satureja montana* and *S. subspicata*, from three sympatric populations along the eastern Adriatic coast have been studied. The individuals from the same locality share the same ecological conditions and observed differences should be due to the genetic variability of two studied species. The surface flavonoids were obtained from 20 (3-5 from each population) individuals by washing surface flavonoids for 5min with MeOH followed by *in vacuo* concentration and filtration with nylon net (0.45 $\mu$ m). The composition was analysed by HPLC-MS and HPLC-DAD. Three solvents in gradient were used: ddH<sub>2</sub>O, MeOH and 1% FA in ACN, under the constant flow rate of 1ml/min. Eleven flavonoids were detected. 5,6-diOH-7,3'4'-triMe flavone, was the dominant compound in *S. montana* individuals, while xanthomicrol dominated the surface flavonoids of *S. subspicata*. The flavonoid profile of *S. subspicata* shows a dominance of one component with more than 50%. The total number of compounds found in *S. montana* populations was eight while in *S. subspicata* it varied from seven to nine. Based on univariate (ANOVA) and multivariate analyses (PCA, CDA, HCA), in all three localities, individuals belonging to the same taxon grouped closer and separated significantly from individuals belonging to the other taxon (species). Chemotaxonomic and ecological implications are further discussed.

**KEYWORDS:** *Satureja subspicata*, *S. montana*, sympatric populations, surface flavonoids, chemotaxonomy



Oral presentation 05 06 39

**PHYTOCHEMICAL SCREENING AND EVALUATION OF ST. JOHN'S WORT (*HYPERICI HERBA*, *HYPERICUM PERFORATUM*, HYPERICACEAE) BIOLOGICAL POTENTIAL – THE IMPORTANCE OF CRUDE DRUG QUALITY CONTROL**

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St John's wort is a medicinal plant widely used in traditional, as well in conventional medicine. Considering the rising market demand for *Hyperici herba* as a result of its medical potential, quality control of crude drug is of high importance. The aim of this study was chemical profiling of *Hyperici herba* samples as a part of quality assessment, followed by evaluation of biological potential. Chemical profiling of *Hyperici herba* samples obtained at local markets, pharmacies and health food stores of Balkan Peninsula countries, Austria and Turkey was performed by HPLC-DAD. Furthermore, the water alcoholic extracts of the collected samples were evaluated in the aspect of antioxidant potential, as well as for the ability to inhibit biologically important enzymes such as acetylcholinesterase (AChE), monoamine oxidases A and B (MAO-A, MAO-B), α-amylase and α-glucosidase. High variability in the amounts of hypericin, hyperforin, rutin, quercetin, gallic, chlorogenic, caffeic and *p*-hydroxybenzoic acid within samples was noticed. Generally, the extracts exhibited significant potential to inhibit MAO-A and α-glucosidase, which supports the proved antidepressant potential of this drug, but also points toward a possibility of its use in diabetes type 2 treatment. The results of antioxidant potential evaluation suggest strong potential to neutralize hydroxyl and nitroso radicals, but moderate inhibition potential of lipid peroxidation process. Overall, the conducted study emphasizes the importance of crude drug quality control and identifies some of the primary factors which significantly affect the tested parameters.

**KEYWORDS:** *Hyperici herba*, Hypericaceae, quality control, HPLC-DAD, enzyme inhibition, antioxidant

Oral presentation 06 06 47

**CYPSELAE FATTY ACID COMPOSITION OF TWO SUBSPECIES OF BALKAN AMPHORICARPOS AUTARIATUS BLEČIĆ & MAYER (COMPOSITAE)**

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The genus *Amphoricarpos* Vis. (Compositae-Cardueae-Carduinae) belongs to the *Xeranthemum* group, which, based on plastid and nuclear analysis, represents a natural group, well characterized on molecular and morphological features. *Amphoricarpos* species are heterocarpic perennial chasmophytic plants, mountain endemics in the eastern Mediterranean (the Balkans, Anatolia and the Caucasus). Taxonomy of the genus *Amphoricarpos* is complex and ambiguous. According to Blečić and Mayer there are three taxa distributed on the Balkan Peninsula: *A. neumayerianus* (Vis.) Greuter (*A. neumayeri* Vis.), *A. autariatus* ssp. *autariatus* Blečić & Mayer and *A. autariatus* ssp. *bertisceus* Blečić & Mayer. Some authors have suggested that all the Balkan populations should be treated as a single species – *A. neumayerianus* (Vis.) Greuter. We analyzed fatty acid composition of central and outer cypselae of two subspecies of *A. autariatus*, using gas chromatography coupled with flame ionization detector (GC-FID). Out of 8 fatty acids detected, 6 were identified, ranging from palmitoleic (C16:0) to stearic (C18:0) acids. Linoleic acid was dominant in the central, as well as in the outer cypselae of both studied taxa, ranging from 31.8 to 57.67%. Unsaturated fatty acids were predominant, ranging from 62.43 to 81.58%. Fatty acids profile differs among studied taxa, as well as between central and outer cypselae of the same taxon. As cypselae fatty acids composition may be valuable as taxonomic marker at infraspecific level, our results demand further examination of all Balkan *Amphoricarpos* taxa from a phytochemical and chemotaxonomic point of view. Moreover, the rest of the *Amphoricarpos* taxa from Georgia and Turkey should be included in the analysis, with the main goal of contribute to the phylogeny of this small, but very complex genus.

**KEYWORDS:** *Amphoricarpos*, Asteraceae, fatty acids, phytochemistry

Oral presentation 07 06 31

**THE INFLUENCE OF *SATUREJA KITAIBELII* WIERZB. EX HEUFF. ADDITION ON THE OXIDATIVE STABILITY OF SUNFLOWER AND OLIVE OIL DURING STORAGE**

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Oxidative stability is an important parameter of edible oils quality. Autoxidation of oils results in production of various products which significantly affect the sensory characteristics of oil, as well as their nutritional value. Antioxidants have the ability to slow down the oxidation process, while currently the preference is given to those obtained from natural sources. The aim of the research was time-dependent monitoring of parameters of oxidative degradation in sunflower and olive oil with the addition of *Satureja kitaibelii* by chemical and physical methods. The content of primary oxidation products was determined by peroxide value method and evaluation of conjugated dienes and trienes, while the content of secondary oxidation products was determined by evaluation of thiobarbituric acid reactive substances (TBARS). The obtained results indicated that peroxide values and levels of thiobarbituric acid reactive substances were lower in samples with added *Satureja kitaibelii*, while the levels of conjugated dienes and trienes were not affected by the addition of herb. Addition of *Satureja kitaibelii* significantly slows the lipid oxidation process in olive and sunflower oil samples during storage time. Also, *Satureja kitaibelii* might represent a potential source of natural antioxidants for aromatization of edible vegetable oils in the future.

**KEYWORDS:** *Satureja kitaibelii*, oxidative stability, sunflower oil, olive oil

Oral presentation 08 06 59

**POTENTIAL OF *MELISSA OFFICINALIS* POSTDISTILLATION WASTE EXTRACTS - PHARMACODYNAMIC STUDIES**

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Lemon balm (*Melissa officinalis* L., Lamiaceae) leaf is mainly used for isolation of essential oil which has numerous phar-

macological activities. Since essential oil is present in low amounts in the leaf, large quantity of plant material remains unused. Recently, investigation of various lemon balm leaf extracts has been given a great attention. In this study, chemically well characterised ethanol extracts of lemon balm leaf, before and after hydrodistillation, were evaluated for their antioxidant and anticholinesterase activity *in vitro* and potential anxiolytic and antidepressive activity, influence on motor coordination and memory on experimental animals, as well as possible interaction with applied conventional drugs. *In vitro* experiments were conducted spectrophotometrically on several antioxidant tests and following Ellman method for evaluation of influence on acetylcholinesterase activity. Pharmacodynamic studies were performed using rotarod performance, tail suspension, elevated plus maze and novel object recognition tests on Swiss Albino male mice. Both examined extracts expressed notable antioxidant and anticholinesterase activity. None of examined extracts exhibited influence on motor activity nor antidepressive effect. Both standard and deodorised leaves extracts of lemon balm showed significant anxiolytic effect, but in applied dosing regimen influence on memory of experimental animals was not recorded. Moreover, there was no interactions with applied antidepressive, anxiolytic and sedative drugs. These findings indicate that postdistillation waste remaining after hydrodistillation of essential oil of lemon balm could be used as a potential source for pharmaceutical industry due to its anxiolytic and anticholinesterase activity.

**KEYWORDS:** lemon balm, postdistillation waste, antioxidant, acetylcholinesterase, anxiolytic effect, antidepressive activity, motor activity, memory

Poster presentation 09 06 58

**ANATOMY AND COMPOSITION OF THE ESSENTIAL OIL OF *LARIX DECIDUA* L. (PINACEAE)**

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*Larix decidua* L. is a deciduous coniferous native to the mountains of central Europe, in the Alps and Carpathians. The aim was to investigate anatomy and the chemical composition of the essential oils of needles, stems and strobilus of *L. decidua*. The samples were collected from larch trees from cultivated conditions (Belgrade, Serbia.) and from natural habitat (Alps, Austria). Needles of *L. decidua*, on cross section are more or less elliptically shaped. Two small secretory canals are in the mesophyll of the needles, on their lateral sides, just below the epidermis. The young stem (primary structures) have canals

in cortex, some are just below the epidermis and many, that are in forming phase above the phloem. The older stem (perennial stem) has many secretory canals in the parenchyma of cortex and in the xylem of secondary wood. The essential oils were obtained by hydrodistillation and qualitative and quantitative analysis was performed by GC-FID and GC-MS. Small amounts of essential oils were extracted from the needles (0.06-0.41%), stems (0.31-0.54%) and strobilus (0.16 % and 0.19 %) of *L. decidua*. The monoterpene (28.70-50.40%) and sesquiterpene compounds (41.80-67.20%) dominated in the essential oil of needles, monoterpenes (80.50-91.70%) in the oil of stems, diterpenes (46.50% and 40.50%) and monoterpenes (24.20% and 22.20%) in the oil of strobilus. Main compounds in the essential oil of needles represented germacrene D (15.2-46.9%),  $\delta$ -3-carene (4.6-24.6%) and  $\alpha$ -pinene (6.3-14.1%), in the oil of stems  $\alpha$ -pinene (12.8-21.8%),  $\delta$ -3-carene (8.6-14.3%) and sabinene (0.5-10.3%), while in the oil of strobilus diterpene of abietane type (4.1-10.2%) and nor-abietatriene (3.9-9.2%) were the most abundant. The larch needles and stem essential oils from cultivated and natural habitats were quite similar in qualitative composition. However, the essential oil of needles from natural habitat contained higher content of germacrene D and  $\alpha$ -pinene and the oil of stem  $\alpha$ -pinene and trans-verbenaol. The oil of larch strobilus from natural habitat was characterized by manool and 3- $\alpha$ -acetoxy-manool, diterpenes that were not present in the oil of cultivated sample.

**KEYWORDS:** *Larix decidua*, anatomy, needles, stem, strobilus, essential oil

Poster presentation 10 06 53

### PIMPINELLA TRAGIUM VILL. (APIACEAE) – ANATOMY AND ESSENTIAL OIL COMPOSITION

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*Pimpinella tragi* Vill. is a perennial, herbaceous plant inhabiting dry grasslands, limestone rocky areas and screes of wide sense Mediterranean area, east European lowlands and Caucasus. This study explores the anatomical structure, content and composition of the essential oil of the vegetative organs and fruit of this species. The plant material was collected on two localities: the village Izvor (Bosilegrad, Serbia) and Mt. Galičica (FYR Macedonia). The anatomical studies were conducted on permanent slides obtained by standard

method of preparation for viewing under a light microscope. The essential oils obtained by hydrodistillation from roots (collected during flowering and fruiting period), aerial parts with inflorescences, and fruits, were analysed by GC-FID and GC-MS. The anatomical analysis revealed a secondary structure of root and primary structure of stem with closed collateral vascular bundles. Leaves are isobilateral, amphistomatic; petiole is with arched vascular bundles and the fruit (mericarp) is semi-circular and lightly ribbed in cross section. Non-glandular unicellular and bicellular cuneate trichomes are sparsely distributed on the stem, leaves and petiole, but very dense on the fruit. Secretory channels are present in all organs: in the parenchyma of the root and stem cortex, stem pith, in the phloem of root and vascular bundles of stem and petiole, by the leaf vascular bundles and in the fruit pericarp. Essential oil yields from different parts of *P. tragi* ranged from 0.2-1.1% (v/w). The main compounds in the oils from roots (in both stages, from both localities), as well as from aerial parts and fruits from Mt. Galičica are C-12 norsesquiterpenes (trinorsesquiterpenes): pregeijerene (29.0-56.2%) and gejerene (14.1-22.9%), whereas those from aerial parts and fruits from village Izvor are  $\beta$ -bisabolene (19.1-57.2%) and a phenylpropanoid epoxy-pseudoisoeugenyl-2-methoxybutyrate (17.4-22.2%). Trinorsesquiterpenes, which were also found to be characteristic constituents in some other *Pimpinella* species oils, are the most dominant constituents in all investigated oils, except in the oil of fruit from village Izvor. In addition, phenylpropanoids of pseudoisoeugenol type are present in all oils confirming them as chemical markers of *Pimpinella* species analysed so far.

**KEYWORDS:** *Pimpinella tragi*, anatomy, secretory canals, essential oil, C-12 norsesquiterpenes, phenylpropanoids

Poster presentation 11 06 29

### CHEMOSYSTEMATIC EVALUATION OF THE COMPOSITION OF LEAF AND FLOWER ESSENTIAL OILS OF EIGHT HERACLEUM L. TAXA FROM SOUTHEASTERN EUROPE

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The genus *Heracleum* L. (Apiaceae) is represented by a dozen of native sub(species) in Southeastern Europe. In this study, the composition of the essential oils obtained from the leaves and the flowers of eight taxa of this genus, collected in Serbia, Montenegro, Macedonia and Slovenia, was statistically analyzed to evaluate its chemosystematic significance. Investigated taxa included *H. orphanidis* Boiss. and the representatives of *H. sphondylium* group: *H. sphondylium* L., *H. sibiricum* L., *H. montanum* Schlecht. ex Gaudin, *H. ternatum* Velen., *H. pyrenaicum* subsp. *pollinianum* (Bertol.) F. Pedrotti & Pignatti

ti, *H. pyrenaicum* subsp. *orsinii* (Guss.) F. Pedrotti & Pignatti and *H. verticillatum* Pančić. Essential oils were hydrodistilled using Clevenger-type apparatus and analyzed by GC-FID and GC-MS. Chemosystematic significance of their components was evaluated using multivariate statistics: principal component analysis (PCA), non-metric multidimensional scaling (nMDS) and unweighted pair-group arithmetic averages clustering (UPGMA). The analyses included our previously published data on the oils of eight samples of the leaves and three of the flowers, as well as additionally analyzed oils of eight samples of the leaves and five of the flowers. Leaf and flower oils of investigated members of *H. sphondylium* group were dominated by various sesquiterpenes [(*E*)-caryophyllene, (*E*)-nerolidol, (*E*)- $\beta$ -farnesene,  $\alpha$ -trans-bergamotene, germacrene D,  $\beta$ -bisabolene and/or  $\beta$ -sesquiphellandrene], phenylpropanoids [apiol, methyl eugenol, elemicin and/or (*Z*)-isoelemicin], and/or monoterpene limonene. On the other hand, leaf and flower oils of *H. orphanidis* were rich in aliphatic esters, mostly octyl acetate. Separate statistical analyses of the compositions of the leaf oils and the flower oils demonstrated segregation of *H. orphanidis* from investigated representatives of *H. sphondylium* group, and grouping of the subspecies of *H. pyrenaicum* within this group. Morphologically related species *H. sibiricum* and *H. ternatum* were closely located in PCA and nMDS, and in UPGMA even shared the same cluster. PCA showed that some of both aforementioned dominant constituents and those present in lower amounts influenced the separation of investigated taxa. It can be concluded that applied multivariate statistical methods demonstrated the grouping of investigated *Heracleum* taxa according to their current systematics, and justify further similar study on the essential oils of more species of this genus.

**KEYWORDS:** *Heracleum* taxa, leaf and flower essential oils, GC-FID and GC-MS, PCA, nMDS, UPGMA

Poster presentation 12 06 30

### DPPH RADICAL SCAVENGING POTENTIAL OF THE ROOT ESSENTIAL OILS OF FIVE HERACLEUM L. TAXA

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In the Balkan Peninsula, *Heracleum* L. taxa (Apiaceae) were traditionally used for the treatment of various digestive and respiratory diseases, epilepsy, hypertension and sexual weakness. The purpose of this work was to investigate 2,2-diphenyl-1-picrylhydrazyl (DPPH) radical scavenging ability of the root essential oils of some of the Balkan *Heracleum* taxa, i.e. *H. sibiricum* L., *H. ternatum* Velen., *H. verticillatum* Pančić,

*H. pyrenaicum* subsp. *pollinianum* (Bertol.) F. Pedrotti & Pignatti and *H. pyrenaicum* subsp. *orsinii* (Guss.) F. Pedrotti & Pignatti. GC-FID and GC-MS analysis of these oils, obtained by hydrodistillation using Clevenger-type apparatus, revealed the domination of monoterpenes, mostly  $\beta$ -pinene (26.2-47.3%). Additionally, *H. sibiricum* root oil was rich in phenylpropanoids, mainly elemicin (25.6%) and methyl eugenol (22.3%). In colorimetric DPPH assay, the strongest activity was exhibited by *H. sibiricum* oil ( $SC_{50}$ =5.19  $\mu$ L/mL), followed by *H. pyrenaicum* subsp. *orsinii*, *H. ternatum*, *H. pyrenaicum* subsp. *pollinianum* and *H. verticillatum* oils ( $SC_{50}$ =7.85-12.33  $\mu$ L/mL). In TLC-DPPH (dot-blot) test, three the most active root oils, i.e. those of *H. sibiricum*, *H. ternatum* and *H. pyrenaicum* subsp. *orsinii* revealed yellow anti-DPPH zones ( $R_f$ =0.30-0.42), which were then eluted and analyzed by GC-FID and GC-MS. It was shown that elemicin and methyl eugenol, dominant in *H. sibiricum* oil, were also the most abundant in its anti-DPPH zone (64.5 and 19.5%).  $\beta$ -Pinene and other monoterpene hydrocarbons were not detected in this, and also in the active zones of *H. ternatum* and *H. pyrenaicum* subsp. *orsinii* oils (both contained two closely located anti-DPPH zones, which were eluted together). Anti-DPPH zones of *H. ternatum* oil were dominated by trans-sabinol (21.8%), spathulenol (21.7%) and (*E*)-sesquivalandulol (13.1%), and those of *H. pyrenaicum* subsp. *orsinii* oil by (*E*)-sesquivalandulol (14.5%) and intermedeol (13.6%). These oxygenated terpenes were detected in *H. ternatum* and *H. pyrenaicum* subsp. *orsinii* oils only in small quantities. Another minor constituent of *H. pyrenaicum* subsp. *orsinii* root oil, (*Z*)-falcarinol, was among the dominant ones in its anti-DPPH zones (11.6%).

**KEYWORDS:** *Heracleum* taxa, root essential oils, GC-FID and GC-MS, DPPH radical, phenylpropanoids, oxygenated terpenes

Poster presentation 13 06 36

### CHEMICAL COMPOSITION OF ESSENTIAL OIL OF ENDEMIC SPECIES ACINOS ORONTIUS FROM BOSNIA AND HERZEGOVINA

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The genus *Acinos* belongs to the family Lamiaceae, and represented by ten species native to southern Europe and western Asia. Its name comes from the Greek word *akinos*, the name of a small aromatic plant. *Acinos orontius* (K. Malý) Šilić is a synonym of *Clinopodium alpinum* subsp. *orontium* (K. Malý) Govaerts, and is endemic species in Bosnia and Herzegovina. The aim of this work was to determine content and composi-

tion of *A. orontius* essential oil. Essential oil, almost odourless, was isolated by hydrodistillation, in low yield (0.08%). Coupled gas chromatography–mass spectrometry (GC–MS) technique was used to characterize its volatile profile. Fifty seven components were identified comprising 84.3% of total oil, and thirty five of these compounds were in very low content, marked as trace compounds. The main constituents were alkanes (39.2%) with pentacosane (12.0%) and hexacosane (11.4%) as the major components. Oxygenated sesquiterpenes caryophyllene oxide (7.7%) and spathulenol (5.8%) were found as additional significant constituents of the essential oil. To the best of our knowledge, this is the first study reporting chemical composition of essential oil of *A. orontius* from BiH.

**KEYWORDS:** *Acinos orontius*, chemical composition, essential oil, GC-MS

Poster presentation 14 06 37

### VOLATILE CONSTITUENTS OF *MELALEUCA ALTERNIFOLIA* ESSENTIAL OIL

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Tea tree is the common name for *Melaleuca alternifolia*, a tree of the family Myrtaceae, native to the subtropical coastal regions of New South Wales and Queensland, Australia. Essential oil of this plant has been used for almost 100 years in Australia but is now available worldwide. Tea tree oil has antiseptic and anti-inflammatory activity and is used in skin and oral hygiene products. Commercial available essential oil of *Melaleuca alternifolia*, as well as headspace of the oil were analyzed via GC-MS. Both samples were characterized with high percentage of oxygenated monoterpenes, i.e. 48.2% and 40.5% respectively. The principal constituents in the essential oil were terpinene-4-ol (38.0%) and  $\gamma$ -terpinene (15.9%), while the main components of its headspace were terpinene-4-ol (31.7%) and *p*-cymene (26.1%). In addition, the headspace analysis of a cosmetic product, skin cream, with tea tree oil as one of the components, was performed. In comparison to headspace of the oil, significant differences in content and composition were recorded. Predominate compounds were oxygenated monoterpenes 47.1%, but the main constituent was 2-phenoxyethanol (34.5%) which is used as a preservative in cosmetic products and also as a stabilizer in perfumes and soaps.

**KEYWORDS:** *Melaleuca alternifolia*, essential oil, headspace, GC-MS

Poster presentation 15 06 54

### VARIATION OF IMORTELLE (*HELICHRYSUM ITALICUM* (ROTH) G. DON) ESSENTIAL OIL COMPOSITION DEPENDING ON METHOD OF PROCESSING AND TIME OF HARVEST

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Immortelle (*Helichrysum italicum* /Roth/ G. Don) is a perennial subshrub that belongs to Asteraceae family, widespread in the Mediterranean. It is used in cosmetic and pharmaceutical preparations due to its medicinal properties. In recent years immortelle has become the most economically valuable medicinal plant in Croatia. It is both wild harvested and grown commercially. In order to obtain essential oil with desirable composition, which meets commercial demands, it is necessary to investigate the optimal method of processing immortelle as well as the optimal harvesting time. We conducted a preliminary study on a small number of samples to determine the differences in chemical composition of essential oils obtained from fresh and dried biomass. The investigations were carried out on four natural populations of immortelle from Croatia. The essential oil was obtained by hydrodistillation, and its content and composition was analysed by gas chromatography and mass spectrometry GC-MS. The results showed differences in essential oil content and composition between the fresh and dried biomass. To determine the influence of the harvest time on the essential oil composition, immortelle essential oil was analysed by GC-MS in the plants shortly before flowering and in the stage of full blossom. The results showed that the stage of harvest plays important role in variation of essential oil composition in immortelle.

**KEYWORDS:** *Helichrysum italicum*, essential oil composition, harvesting time, fresh and dry biomass, GC-MS

Poster presentation 16 06 55

### VARIABILITY OF ESSENTIAL OIL OF DIFFERENT POPULATIONS OF *TEUCRIUM MONTANUM* L. (LAMIACEAE) FROM BALKAN PENINSULA

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*Teucrium montanum* L. is widely distributed in the Balkan Peninsula, extending from the sea coast to altitudes of over 2100 m a. s. l. with the highest number of occurrences in the zone between 500 and 1000 m. The aim of the study was to investigate the chemical composition of essential oil of aerial parts of *T. montanum* from 14 different populations from Balkan Peninsula, ten from Serbia (SR-Trešnjica canyon, SR-Brdjanska gorge, SR-Goč, SR-Gornjak gorge, SR-Grza canyon, SR-Jelašnica, SR-Kopaonik, SR-Maglič, SR-Rtanj, SR-Sićevačka gorge), two from Greece (GR-Olimp, GR-Ossa) and two from Albania (AL-Deja, AL-Skadar). The essential oils were obtained by hydrodistillation and qualitative and quantitative analysis was performed by GC-FID and GC-MS. The aerial parts of *T. montanum* contained only small amounts of oil (traces-0.7%), predominantly composed by sesquiterpene compounds (58.0-99.0%). One population from Albania (Deja), two from Greece (Olimp, Ossa) and two from Serbia (Sićevačka gorge, Trešnjica canyon) contained more than 90% of sesquiterpenes. The composition of essential oils was quite variable and the main compounds in almost all oils were germacrene D (trace-45.5%), sabinene (trace-23.1%),  $\alpha$ -pinene (trace-20.7%), limonene (trace-20.4%), (*E*)-caryophyllene (2.9-14.5%),  $\gamma$ -cadinene (trace-13.8%) and  $\delta$ -cadinene (trace-12.0%). The cluster analysis revealed the separation of investigated essential oils to five clusters. The first cluster (AL-Skadar, SR-Brdjanska gorge, SR-Gornjak gorge, SR-Grza canyon, SR-Goč) is characterized by high germacrene D content (17.6-45.5%), the second (SR-Maglič, SR-Kopaonik, SR-Sićevačka gorge) by high sesquiterpene alcohol shyobunol content (14.6-55.2%), the third (AL-Deja, SR-Jelašnica) by high  $\alpha$ -bisabolol content (31.2%-43.7%) and shyobunol (3.2%) in population AL-Deja, the fourth (SR-Rtanj) by high *cis*-sesquisabinene hydrate content (42.4%) and the fifth (GR-Olimp, GR-Ossa, SR-Trešnjica canyon) by high germacrene D-4-ol (5.96%-61.9%), and  $\beta$ -eudesmol (14.9%) and *epi*- $\alpha$ -cadinol (10.2%) content in GR-Olimp population. Such a high variability needs more research to define relationships among populations.

**KEYWORDS:** *Teucrium montanum*, aerial parts, essential oil, GC-FID, GC-MS, variability

Poster presentation 17 06 10

### SOLUBLE SUGAR COMPOSITION OF ACHENES IN ENDEMIC *TRIPLEUROSPERMUM CALLOSUM* AND *T. MONTICOLUM* (ASTERACEAE) FROM TURKEY AND IT'S ECOLOGICAL IMPLICATIONS

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Soluble sugars play an important role in desiccation tolerance in the seeds of various species as well as representing potential food sources for organisms. This study performs the evaluation of soluble sugar (glucose, fructose, sucrose and maltose) composition in achenes of two endemic *Tripleurospermum* (Asteraceae) species (*T. callosum* and *T. monticolum*) from Turkey using a high performance liquid chromatography (HPLC) system equipped with a refractive index detector (IR). The presence of glucose, fructose, maltose and sucrose in the samples were identified using the Chemstation software and authentic external sugar standards. The composition of the soluble sugars of these species growing on the steppes in Turkey is presented here for the first time. Among the soluble sugars identified in both species, the sucrose content is higher than that of other soluble sugars, while the maltose content is lower than other soluble sugars. The ecological significance of the soluble sugars of the achenes in these species is also discussed.

**ACKNOWLEDGEMENTS:** This research was supported by the Scientific and Technological Research Council of Turkey (Tubitak, project no. 106T162).

**KEYWORDS:** *Tripleurospermum*, endemic, soluble sugar, HPLC, Turkey

## DIFFERENCES IN LEAF CUTICULAR WAXES AMONG *PINUS HELDREICHII*, *PINUS NIGRA* AND *PINUS PEUCE*

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Fresh needles were collected from natural populations of 1) *Pinus heldreichii* (Mt. Lovćen, Mt. Zeletin, Mt. Bjelasica, and from isolated localities (Mt. Zlatibor to Mt. Pešter plateau), 2) *Pinus nigra* (Mt. Tara-Banjska Stena, Mt. Tara-Omar, Mt. Tara-Zmajevački potok, Priboj-Crni vrh, Mt. Goč-Gvozdac, Mt. Dukat-Jarešnik, and Lazareva reka Canyon-Kovej) and 3) *Pinus peuce* (Mt. Zeletin, Mt. Sjekirica and Mt. Mokra Gora). The total wax of each sample was extracted by immersing 3 g of needles in 10 ml of *n*-hexane for 45 s. GC and GC/MS analyses were carried out with an Agilent 7890A apparatus equipped with an inert 5975C XL EI/CI mass spectrometer detector (MSD) and flame ionisation detector (FID), connected by capillary flow technology 2-way splitter with make-up. A HP-5MS capillary column (30 m x 0.25 mm x 0.25 µm) was used. The components were identified based on their retention index and comparison with reference spectra (Wiley and NIST databases), as well as by the retention time locking (RTL) method and the RTL Adams database. In *P. nigra* *n*-alkanes ranged from C<sub>16</sub> to C<sub>33</sub>, while in *P. peuce* and *P. heldreichii* ranged from C<sub>18</sub> to C<sub>33</sub>. The most abundant *n*-alkanes in *Pinus nigra* was pentacosane (C<sub>25</sub>) (mean value ± SD: 19,71±4,92), in *Pinus peuce* was nonacosane (C<sub>29</sub>) (15,54±4,39), and in *Pinus heldreichii* was tricosane (C<sub>23</sub>) (12,21±3,01). Nonacosane was more abundant in *Pinus peuce* (15,54±4,39) and *Pinus nigra* (10,17±0,26) than in *Pinus heldreichii* (9,09±2,05). On the other way, *Pinus heldreichii* and *Pinus nigra* had abundant pentacosane (C<sub>25</sub>) (10,79±1,51 and 19,71±4,92, respectively). Furthermore, *P. heldreichii* had the most abundant eicosane (C<sub>21</sub>) (9,18±3,50) and docosane (C<sub>22</sub>) (8,44±3,65). It seems that the greatest distance is among *P. heldreichii* and *P. peuce*. It is in accordance with taxonomic classification, where two-needle pines, *P. nigra* and *P. heldreichii*, belong to subgenus *Pinus* (Diploxylon or hard-pines), while *P. peuce*, five-needle pine, belongs to subgenus *Strobos* (Haploxylon or soft pines). Presented results need further studies at population level and use of multivariate statistical analyses in order to get better insight into usefulness of *n*-alkanes in ecological, geographical and/or taxonomic differentiation of pine species.

**KEYWORDS:** *Pinus*, *n*-Alkanes, waxes, needles

## NEEDLE *N*-ALKANES, PRIMARY ALCOHOLS AND DITERPENES: APPLICATION TO THE ANALYSIS OF POPULATION DIFFERENTIATION IN *PINUS MUGO* TURRA

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*Pinus mugo* Turra *sensu stricto* (dwarf mountain pine) is a conifer species with subalpine character and highly fragmented range, occurring in the mountain massifs of Central, Southern, and Eastern Europe. In this paper, phytochemical diversity and differentiation of eight *P. mugo* natural populations from the Julian Alps, Southern Carpathians and Balkan Peninsula were analyzed in regard to epicuticular wax compounds. GC-MS (Gas Chromatography-Mass Spectrometry) and GC-FID (Gas Chromatography-Flame Ionization Detector) analyses of *n*-hexane extracts of needle samples, collected from 111 individuals of *P. mugo*, have revealed the presence of five diterpenes, three primary alcohols and 11 *n*-alkanes ranging from C<sub>19</sub> to C<sub>29</sub>. *n*-Nonacosane (C<sub>29</sub>) was the most abundant compound at the species level, but significant variations among the populations in the contents of most of the detected compounds were observed. The multivariate statistical analyses (Canonical Discriminant Analysis and Agglomerative Hierarchical Clustering) suggested existence of two chemical entities: the Alpine and the South Carpathian, while the Balkan populations appeared heterogeneous as two of them belonged to the Alpine and one to the South Carpathian group. Obtained results are largely consistent to those previously published in reference to morphological and molecular characters of *P. mugo*, supporting the hypothesis of colonization of Balkan Peninsula from two different glacial refugia: from the Alps (across the Dinarides) and from the Southern Carpathians. According to simple linear regression, contents of epicuticular wax compounds have shown generally weak correlations with tested environmental factors, leading to the conclusion about their genetic conditioning. Therefore, the significant chemical structuring between populations of *P. mugo* from Alps and Southern Carpathians is likely to be explained by an ancient fragmentation and long-term isolation, causing independent genetic process and different demographic events.

**KEYWORDS:** *Pinus mugo*, chemodiversity, epicuticular wax, Alps, Carpathians, Balkans

## PHENOLIC PROFILE AND ACETYLCHOLINESTERASE INHIBITORY ACTIVITY OF *PLANTAGO ARGENTEA* CHAIX.

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Genus *Plantago* comprises more than 250 cosmopolitan species, which are commonly used both in nutrition and traditional medicine. But, chemical composition and biological activity of only several *Plantago* species, such as *P. lanceolata* L. and *P. major* L. are well studied. About other species, such as *P. argentea* Chaix., there are only few data in literature. Since polyphenolic compounds are responsible for many biological activities of plants, and, potentially, could be used as chemotaxonomic markers, the aim of this study was to investigate polyphenolic profile of water and methanolic extracts of *P. argentea*. Furthermore, inhibitory potential of *P. argentea* towards acetylcholinesterase, an enzyme which is deeply involved in pathogenesis of neurodegenerative disorders, was investigated. In order to thoroughly evaluate polyphenolic profile of extracts, quantitative analysis of 47 polyphenolics, including 16 phenolic acids, 26 flavonoids, 3 coumarins and 2 lignans, was performed using the LC-MS/MS technique. The Ellman's spectrophotometric method, adapted to 96-well microplates, was used to determine inhibitory activity of extracts towards acetylcholinesterase. LC-MS/MS analysis of selected polyphenols resulted in 21 detected from 47 investigated compounds and, in general, slightly lower polyphenolic content in water extract. Among analyzed polyphenolics, luteolin was dominant flavonoid in both extracts, while ursolic acid was the most frequent phenolic acid. Considering neuroprotective activity, water extract (IC<sub>50</sub> = 1.34 mg/mL) evinced better potential of acetylcholinesterase inhibition than methanolic (IC<sub>50</sub> = 3.01 mg/mL). However, in comparison to physostigmine (IC<sub>50</sub> = 0.66 µg/mL), a well known acetylcholinesterase inhibitor, both extracts manifested modest activity. This study reports novel and valuable data about polyphenolic profile and neuroprotective activity of *P. argentea*. Moreover, obtained results support further studies of chemical composition and biological activities of examined extracts, in order to determine its potential use in food and pharmaceutical industry.

**KEYWORDS:** *Plantago argentea* Chaix, phenolic profile, acetylcholinesterase

## PHENOLIC CONTENT AND BIOLOGICAL ACTIVITIES OF ANISE SEED EXTRACTS

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The aims of this work were to determine (i) the total phenolic content of extracts of anise (*Pimpinella anisum* L.); (ii) the antioxidant activity of the samples by means of four different antioxidant methods; (iii) the effectiveness of the samples on inhibition of the growth of bacteria strains; (iv) inhibitory activity of extracts against butyrylcholinesterase (BChE). Six extracts of *P. anisum* were prepared using Soxhlet, ultrasonic and reflux extraction with three different solvents (methanol, water, and *n*-hexane). The total phenol content was determined using Folin-Ciocalteu's reagent. Radical-scavenging and antioxidant activity were tested by means of 1,1-diphenyl-2-picrylhydrazyl (DPPH) method; 2,2'-azino-bis(3-ethylbenzothiazoline-6-sulphonic acid) (ABTS) method; reducing power (RP) method and oxygen radical absorbance capacity (ORAC) tests. The extracts were tested against *Bacillus subtilis*, *Staphylococcus aureus*, *Salmonella enterica*, and *Escherichia coli* using the agar diffusion method. The inhibition of BChE was assessed by modified spectrophotometric method of Ellman. Total phenolic content ranged from 6.44±1.12 mg GAE/g of extracts for ultrasonic *n*-hexane extract of anise (PAUH) to 47.88±2.48 mgGAE/g extracts for ultrasonic methanol extract (PAUM). Reflux methanol extract (PARM) showed the highest potency in DPPH assay (IC<sub>50</sub> of 2.63±0.09 mg/mL), while Soxhlet methanol extract (PASM) exhibited the highest radical ABTS scavenging activity (IC<sub>50</sub> of 1.09±0.05 mg/mL). The most active sample in RP and ORAC assay was ultrasonic *n*-hexane extract (PAUH) (23.18±1.44 mg/mL and 1.47 mmolTE/g, respectively). Total phenolic content showed a correlation with antioxidant activity. The extracts exhibited considerable inhibitory activity against the tested bacterial strains. Maximum activity of Soxhlet *n*-hexane extract (PASH) was observed against *S. aureus* and Soxhlet methanol extract (PASM) against *S. enterica*. Finally, all extracts showed inhibitory activity against BChE and inhib-

itory concentration, IC<sub>50</sub>, ranged from 0.15±0.01 mg/mL to 0.63±0.04 mg/mL.

**KEYWORDS:** *Pimpinella anisum* L., total phenolic content, antioxidant activity, antibacterial activity, butyrylcholinesterase inhibition

Poster presentation 22 06 27

### COMPARISON OF THE BULGARIAN LICORICE POPULATIONS BY THEIR GLYCYRRHIZIN AND FLAVONOID CONTENTS AND *IN VITRO* PROPAGATION RATE

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*Glycyrrhiza glabra* L. underground organs are a source of the triterpenoid saponin glycyrrhizin and many flavonoids used in medicine for treatment of asthma, renal calculus, ulcer, psoriasis, rheumatism. Licorice propagates mainly by horizontal rhizomes. Glycyrrhizin content varies along the year but literature data are contradictory concerning the best season for root harvest. Bulgarian populations shrank during the last decades and the species was protected by the Biodiversity Act and included in the Red Data Book as “endangered”; cultivation was recommended. As populations differ by the content of the bioactive compounds, *in vitro* plants multiplication from selected population would be the most appropriate way toward commercial cultivation. The study deals with comparison of few survivor Bulgarian licorice populations concerning both *in vitro* propagation potential and bioactive compounds accumulation during flowering and seed-production stages and its possible relations with soil composition. Rhizome segments were gathered from *G. glabra* populations along the Danube River, near the villages of Dolni Vit, Koilovtsi, Beltsov, and Baykal; one commercially important Ukrainian population was used as a referent. Segments were washed, cut, dried at 40°C and grinded to powder. Glycyrrhizin content was determined in methanol extracts by HPLC (UV/VIS detector, LC-85B, Perkin Elmer), controlled with Chromulan software (PIKRON). Total flavonoids were determined spectrophotometrically according to the European Pharmacopeia, then aglycones and glycosides were evaluated by semi-quantitative TLC. Soil mechanical composition and several principal elements were analysed. *In vitro* culture was initiated on MS basal medium, and then subcultured on medium supplemented with 1 mg/l kinetin. Tested licorice populations differed significantly from one another according to their glycyrrhizin contents (P<0.001), the richest one being that near Beltsov (29.6±2.3 mg/g). This population distinguished also with highest *in vitro* propagation rate: 19.3 plantlets per explant for 5 months. Total flavonoids were over 0.2% except for Dolni Vit: 0.04%, where one extra aglycone

was detected as well. All extracts were richer in flavonoid aglycones than in glycosides. Some seasonal fluctuations of bioactive compounds were noticed within populations. Population near Beltsov was selected as a source for further clonal propagation due to its highest glycyrrhizin content and best *in vitro* multiplication rate.

**ACKNOWLEDGEMENTS:** Authors are grateful to “Program for Support of Young Researchers and PhD Students at the Bulgarian Academy of Sciences” (Grant no. 17-16/24.07.2017) for the financial support of the study.

**KEYWORDS:** *Glycyrrhiza glabra* L., medicinal plants, bioactive compounds, cultivation

Poster presentation 23 06 32

### POLYPHENOLIC COMPOSITION OF TWO EUPHRASIA ROSTKOVIANA L. SPECIES FROM DIFFERENT LOCALITIES

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The genus *Euphrasia* represents a large group of hemi-parasitic plant species that are widely used in traditional medicine for treatment and prevention of eye conditions, such as conjunctivitis and ocular inflammation. It is well known that polyphenols are often responsible for various biological activities of plant extracts, including anti-inflammatory activity. Since published data regarding phenolic profile of *Euphrasia* species are very scarce, we investigated polyphenolic composition of hydromethanolic extracts of *Euphrasia rostkoviana* L. from two different locations in Serbia. Sixty six phenolic compounds were analyzed by LC-MS/MS technique. Obtained results showed that the content of phenolic compounds in the plant is high. Among 25 compounds detected in the extracts, the most dominant was flavonoid rutin, exceeding 1229 µg/g DE. Other dominant compounds were p-coumaric acid, caffeic acid, quinic acid, luteolin, luteolin-7-O-glycoside, kaempferol-3-O-glycoside, chrysoeriol, amentoflavone and diosmetin. Quantities of quinic acid (8.512 and 35.22 µg/g DE), luteolin (6.266 and 35.22 µg/g DE), luteolin-7-O-glucoside (3.891 and 15.55 µg/g DE), kaempferol-3-O-glucoside (0.719 and 5.425 µg/g DE) and amentoflavone (0.502 and 4.212 µg/g DE) varied greatly among samples from different locations, possibly due to hemi-parasitic nature of the plant. The anti-inflammatory activity has previously been reported for several polyphenols detected in this study, indicating that these compounds are at least partially responsible for anti-inflammatory properties of the plant, which supports its usage

against inflammatory conditions in traditional medicine. This study pointed out that *Euphrasia rostkoviana* is a rich source of phenolic compounds, thus it can be recommended for further studies of biological activities.

**KEYWORDS:** *Euphrasia rostkoviana* L., polyphenolic composition

Poster presentation 24 06 33

### SEASON AND LOCALITY INFLUENCE ON PHENOLIC CONTENT AND BIOACTIVITY OF ROSEMARY EXTRACTS

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*Rosmarinus officinalis* L. (rosemary) is one of the most famous flavouring and medicinal plant of Lamiaceae family mainly due to presence of essential oil and polyphenolic compounds in its leaves. Rosemary is a perennial herb native to the Mediterranean region and cultivated worldwide, so it could be expected that polyphenolic content and bioactivities of rosemary extracts vary broadly depending on the locality and harvesting time. The objective of this study was to compare phenolic and flavonoid contents and bioactivities of extracts obtained from leaves of plants cultivated in Belgrade and Bogatić (Serbia), and Lastva Grbaljska (Montenegro) collected in March, July and November. The crude extracts were obtained using 70% methanol, 70% ethanol and hot distilled water. All of the 27 extracts were screened at concentration of 500 µg/mL for total phenolic content (TPC), total flavonoid content (TFC), as well as for antioxidant activity (2,2-diphenyl-1-picrylhydrazyl (DPPH), ferric reducing ability of plasma (FRAP) and β-carotene bleaching assays). The extracts showed broad range of variations in TPC (30.38-134.60 mg GAE/g), TFC (5.44-25.54 mg QE/g), DPPH assay (49.35-93.53%), FRAP assay (349.52-1716.10 µmol Fe (II)/g) and β-carotene bleaching assay (58.71-82.51%). Regardless of locality, season and applied assay, the lowest values were obtained for aqueous extracts. Since extracts of Lastva samples harvested in all seasons showed the highest values of TPC, TFC and antioxidant activities, they are subsequently selected for evaluation of antineurodegenerative potential against enzymes acetylcholinesterase (AChE) and tyrosinase (TYR) and of cytotoxic activity using MTT (3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide) assay against HCT-116 cancer cell line. Analyzed samples showed inhibition of AChE and TYR (44.92-91.77% and 47.70-67.74%, respectively), especially aqueous extracts of leaves harvested in November (AChE) and July (TYR). In MTT assay, the strongest cytotoxic activity was obtained for 70% ethanolic and 70% methanolic extracts of sample harvested in July (IC<sub>50</sub> values=8.65 µg/mL and 13.08 µg/mL, re-

spectively). Samples from the Montenegrin coast, regardless of harvesting time, showed the highest polyphenolic content and the strongest bioactivities, probably due to similarities of environmental conditions of cultivation and native habitats of rosemary.

**KEYWORDS:** *Rosmarinus officinalis*, extracts, phenolics, bioactivities, different localities and seasons

Poster presentation 25 06 35

### EFFECT OF SALICYLIC ACID AND METHYL JASMONATE ON PRODUCTION OF PHENOL, FLAVONOID AND PIGMENTS OF MADAGASCAR PERIWINKLE CALLUS

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Madagascar periwinkle (*Catharanthus roseus* L.) produces secondary metabolites such as phenol and flavonoid and it has anti-cancer alkaloids which are important in pharmaceutical industry. Salicylic acid (SA) and methyl jasmonate (MJ) as elicitors can increase production of secondary metabolites in plants. In present study, the effects of SA and MJ on fresh weight, flavonoids, phenolics, and pigments of Madagascar periwinkle callus were investigated in a completely randomized design in *in vitro* conditions. In each experiment, the first factor was the concentration of an elicitor, SA or MJ, in 4 levels of 0, 50, 100 and 200 µM, and the second factor was time at 2 levels of 5 and 10 days, with three replications. Based on obtained results, the highest fresh callus weight was obtained in 100 µM MJ, followed by 100 µM SA at a time interval of 5 days. The interaction between time and SA in flavonoids, phenolics, chlorophyll and carotenoids production was significant. The phenolics, chlorophyll and carotenoids were significantly different in the interaction of time and MJ at 1% level. The highest phenolics content was obtained from 100 µM MJ treatment for 5 days, while the highest amount of flavonoid and anthocyanin were obtained in 50 µM SA. On the other hand, the highest amount of chlorophyll and carotenoids was produced in control treatment in 10 days. Increasing the time from 5 to 10 days reduced the levels of phenolics, flavonoids and anthocyanins, but increased chlorophyll and carotenoids content in the Madagascar periwinkle callus. Based on these findings, SA and MJ as elicitors at 50 and 100 µM concentrations in 5 days can significantly increase the production of phenolics, flavonoids and anthocyanins in the Madagascar periwinkle callus.

**KEYWORDS:** salicylic acid, methyl jasmonate, flavonoids, anthocyanin, chlorophyll, carotenoids

**PHENOLIC AND FLAVONOID CONTENT AND ANTIOXIDANT ACTIVITY OF *GLAUCOSCIADIUM CORDIFOLIUM* (BOISS.) BURTT ET DAVIS ETHANOL EXTRACT FROM DIFFERENT PARTS**

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*Glaucosciadium cordifolium* (Boiss.) Burtt et. Davis (syn. *Siler cordifolium* Boiss.) is a monotypic plant of the family Umbelliferae growing in the Mediterranean region. The aim of this work was to determine the content of total phenolics and flavonoids as well as *in vitro* antioxidant activity in 70% ethanolic extracts prepared from different parts (roots, leaves, flowers and stems) of *Glaucosciadium cordifolium*. The antioxidant activity was evaluated using DPPH, ABTS radical scavenging,  $\beta$ -carotene/linoleic acid system while total polyphenolic and flavonoid content were determined via Folin-Ciocalteu method and aluminum chlorid spectrophotometric method, respectively. The results showed that the flower of *G. cordifolium* contain higher amounts of phenolic (137.52  $\pm$  1.11 mg gallic acid equivalent per gram of extract) and flavonoid compounds (155.40  $\pm$  5.94 mg quercetin equivalent per gram of extract) than other parts and possess moderate antioxidant activity (the IC<sub>50</sub> value of flower extract for DPPH and ABTS radical scavenging were 216.07  $\pm$  1.39  $\mu$ g/mL and 29.26  $\pm$  4.62  $\mu$ g/mL, respectively), which may attributed to a strong free radical scavenging, iron chelating and lipid peroxidation inhibitory activities. It was concluded that the extract of *G. cordifolium* may be a phytochemical source showing antioxidant activity that associate with health benefits.

**KEYWORDS:** *Glaucosciadium cordifolium*, antioxidant activity, phenolic compounds, *in-vitro*

**PHENOLIC CONTENT AND ANTIOXIDANT ACTIVITY OF *CHAEROPHYLLUM AUREUM* L. AND *C. HIRSUTUM* L. EXTRACTS (APIACEAE)**

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The genus *Chaerophyllum* comprises about 40 annual, biennial or perennial species. Some *Chaerophyllum* species are used as vegetables and spices in culinary in France, Austria and Turkey, as well as in food industry. *C. aureum* L. and *C. hirsutum* L. are perennial plants with white or pinkish flowers that reach a height of up to 120 cm. Both species inhabit moist and shady habitats. The objective of this study was to determine total phenolic contents and potential radical scavenging activity of different extracts of aerial parts of *C. aureum* and *C. hirsutum* collected on Mt. Kopaonik (Serbia). Ultrasound assisted extraction and different solvents (water, methanol, ethanol and acetone) were used in extraction process. For quantification of total phenols in tested samples, the Folin-Ciocalteu reagent was used and obtained results were presented as mg of galic acid equivalents (GAE) per g of dry extracts (DE). Antioxidant activity was measured by ABTS assay, where flavonol quercetin hydrate was used as a positive control, and the results were presented as equivalents of ascorbic acid (vitamin C). Obtained data indicated that aqueous extract of *C. aureum* in concentration of 2 mg/mL, was the richest in phenols (226.68  $\pm$  14.04 mg GAE/g of DE), while the lowest content was observed in aqueous extract of *C. hirsutum* (68.66  $\pm$  2.12 mg GAE/g of DE). According to results obtained by ABTS method, the highest scavenging potential possessed aqueous extract of *C. aureum* (2.65  $\pm$  0.01 mg vit. CE /g of DE), which was in line with used quercetin hydrate activity (2.75  $\pm$  0.00 mg vit. CE/g of DE). The lowest effect was recorded for ethanolic extract of *C. hirsutum* (0.37  $\pm$  0.01 mg vit. CE/g of DE). In general, positive correlation between measured total phenolic contents and demonstrated antiradical effect was observed. *C. hirsutum* manifested better antioxidant activity, where aqueous extract was the most promising ABTS scavenging agent with the highest phenolic concentration. Results indicated that tested extracts may have potential application as natural antioxidant.

**KEYWORDS:** *Chaerophyllum hirsutum*, *C. aureum*, extracts, total phenols, antioxidant activity, ABTS

**PHENOLIC CONSTITUENTS, ANTIOXIDANT,  $\alpha$ -AMYLASE AND  $\alpha$ -GLUCOSIDASE INHIBITORY ACTIVITIES OF *PYRUS*  $\times$  *VELENOVSKYI* BARK**

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*Pyrus*  $\times$  *velenovskyi* Dostálek (Rosaceae) [*P. pyrastrer* (L.) Burgsd.  $\times$  *P. spinosa* Forssk.] is a deciduous tree up to 5 m height. Leaf-blades 2.2–4.8  $\times$  1.3–2.7 cm,  $\pm$  glabrate, elliptic, gradually narrowed to a rather long petiole. This natural hybrid plant was described from Bulgaria, and has been also found in Serbia. The objective of this work was to investigate the phenolic profile, as well as *in vitro* antioxidant,  $\alpha$ -amylase and  $\alpha$ -glucosidase inhibitory activities of dried methanol extract from the bark of this tree. The plant material was collected in eastern Serbia (Jelašnička Klisura gorge). In the dried bark, the contents of different classes of phenolics were spectrophotometrically determined: total polyphenols (10.36%), tannins (8.78%), procyanidins (4.21%) and phenolic glycosides (3.54%). After pre-extraction with dichloromethane, powdered bark was extracted with methanol by bimaceration at room temperature. In obtained dried methanol extract, using aforementioned spectrophotometric tests, the contents of total polyphenols (33.42%) and tannins (21.55%) were determined, and by HPLC, arbutin, chlorogenic acid, catechin and procyanidin B2 were identified. Its antioxidant activity, i.e. ferric reducing capacity (FRAP), 2,2-diphenyl-1-picrylhydrazyl (DPPH) and  $\cdot$ OH radical scavenging ability, as well as the inhibition of the enzymes  $\alpha$ -amylase and  $\alpha$ -glucosidase were assessed using corresponding colorimetric assays. Tested dried methanol extract exhibited significant DPPH and  $\cdot$ OH radical scavenging abilities (SC<sub>50</sub> = 6.85 and 12.21  $\mu$ g/mL, respectively), and ferric reducing capacity (10.74 mmol Fe (II)/g of dried extract), as well as the inhibition of  $\alpha$ -amylase (IC<sub>50</sub> = 11.4  $\mu$ g/mL) and  $\alpha$ -glucosidase (IC<sub>50</sub> = 5.48  $\mu$ g/mL).

**KEYWORDS:** *Pyrus*  $\times$  *velenovskyi* bark, phenolics, antioxidant activity,  $\alpha$ -amylase and  $\alpha$ -glucosidase inhibition

**EXUDATE FLAVONOIDS AND PYRETHRINS OF AERIAL PARTS OF *TANACETUM CINERARIIFOLIUM***

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*Tanacetum cinerariifolium* (Trevir.) Sch. Bip. (Asteraceae) is a perennial herbaceous plant, endemic to the east Adriatic coast. The species was first used as an insecticide in Croatia, and later in the rest of the world. The insecticidal action of *T. cinerariifolium* is determined by pyrethrins. These compounds are found in all parts of the plant, but are mainly concentrated in the flower heads. The term pyrethrin refers to the six insecticide active ingredients: Pyrethrin-I, Pyrethrin-II, Cinerin-I, Cinerin-II, Jasmolin-I and Jasmolin-II, but there are many other compounds that also include resinous substances - terpenoids, fatty acids, sesquiterpene lactones, flavonoid aglycones and etc. Exudate (surface, external) flavonoids are aglycones accumulated usually on the surfaces of leaves, flowers, and other tissues, they extracted by glandular trichomes or are extruded through the cuticle. Although the content of pyrethrins in flower heads of *T. cinerariifolium* are well studied the data on content of these compounds in aerial parts are limited, more that to the best of our knowledge there no reports about exudate flavonoids in the species. The aim of present study was to analyze the content of exudate flavonoids and pyrethrins of the aerial part of *T. cinerariifolium*. The samples were collected from a naturally occurring population of the species and from an *ex situ* collection of IBER-BAS. Acetone exudates and hexane fractions of collected plant material of both origins, were comparatively analyzed by GC/MS and TLC. Flavonoid aglycones were identified using co-chromatography with authentic samples on different sorbents: silica-gel, polyamide and cellulose. Luteolin 6-methyl ether, quercetagenin 3,6-dimethyl ether and quercetagenin 3,6,3'-trimethyl ether were detected as main flavonoid aglycones of the studied acetone exudates. Luteolin and scutellarein 6-methyl ether were found in traces. Besides the six main insecticide active ingredients a variety of alkanes were identified in the hexane fractions of studied samples. Pyrethrin-I, Pyrethrin-II and Cinerin-II were present in the largest amount. Differences in the qualitative flavonoid composition and pyrethrin profiles of plant material from naturally occurring population and from an *ex situ* collection of IBER-BAS were not established. The study presents for the first time data for exudate flavonoids of *T. cinerariifolium*.

**ACKNOWLEDGEMENTS:** The authors are grateful to the Program for Support of Young Researchers and PhD Students at the Bulgarian Academy of Sciences (Grant no. DFNP - 17-109/28.07.2017) for the financial support.

**KEYWORDS:** Dalmatian pyrethrum, *Pyrethrum cinerariaefolium* Trev., *Chrysanthemum cinerariaefolium* Bocc, GC/MS, TLC, flavonoid

### IMPACT OF EXTRACTION PARAMETERS ON BIOLOGICAL POTENTIAL AND COMPOSITION OF BILBERRY EXTRACTS

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Bilberry has long tradition of medical use, both conventional and alternative, due to its beneficial effect on human health. As oxidative stress has started to have an emerging role in pathogenesis of various diseases, antioxidant potential of bilberries' extracts has come to focus lately. The aim of this study was to examine how different extraction methods affect the effectiveness of extraction of bioactive compounds from bilberry's fruit, as well as to determine antioxidant activity of the analyzed extracts. Investigated extracts were prepared by methanol maceration method, decoction making and infusion making methods. The total amount of phenols was determined by Folin-Ciocalteu method. Total amount of flavonoids was determined by spectrophotometry analysing flavonoid-metal complex forming. The amount of total anthocyanins was determined spectrophotometrically developing the color of antocyanins with hydrochloric acid solution. Antiradical activity of the analyzed extracts was tested on the synthetically prepared DPPH radical (1,1-diphenyl-2-picrylhydrazyl radical). The highest amount of the total phenols is present in decoction, whereas the highest amount of flavonoids and anthocyanins is present in methanol extract. DPPH test showed significant antioxidant potency of all examined extracts, among which decoction has the highest potency. The extraction conditions affect the bilberry's extract composition and antioxidant activity. Yielded results imply that there is the need for further investigation in order to determine detailed chemical composition and isolate the most active compounds responsible for antioxidant potential.

**KEYWORDS:** bilberry, methanol extract, infusion, decoction, chemical composition, antioxidant activity

### CONTENT OF ANTHOCYANINS IN ARONIA MELANOCARPA FRUIT INFLUENCED BY DIFFERENT SOLVENT SYSTEMS

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Lately, there is an increased interest for berries of *Aronia melanocarpa* fruits because of their potential health effects. Numerous studies have shown its significant impact on protection role against cardiovascular disease, and the reduction of cancer risk. The high biological activity of chokeberry is associated with its chemical composition, rich in different antioxidants, especially polyphenolic components. The dominant polyphenols of chokeberry are anthocyanins and their abundance is approximately 25% of total polyphenols. However, anthocyanins are greatly unstable and susceptible to degradation and polymerization, and therefore their utilization is limited. The aim of this work was to determine total monomeric anthocyanins content (TMAC) and degradation/polymerization of anthocyanins using three different solvent systems: 100% methanol (MeOH), 80% MeOH and 50% ethanol (EtOH). Monomeric anthocyanins were measured by the pH-differential method and the anthocyanin content was expressed as cyanidin-3-glycoside (C3G), while the anthocyanin degradation indices were determined by measuring several absorbencies of samples at different wavelengths, which has been treated with sodium bisulfite and expressed as percent polymeric color (PPC). The obtained results for TMAC were: 1.314 mg/g C3G (100% MeOH), 1.465 mg/g C3G (80% MeOH) and 0.661 mg/g C3G (50% EtOH). Percent of polymeric color for different solvent was 7.52% (100% MeOH), 32.93% (80% MeOH) and 46.41% (50% EtOH), respectively. According to exhibited results, it can be concluded that anthocyanins composition and degradation process of *Aronia melanocarpa* fruit differ and depend on the choice of adequate solvent. Results indicate that the most appropriate solvent for extraction of monomeric anthocyanins was 80% MeOH, while the 100% MeOH solvent demonstrated the greatest protective properties against the degradation of the anthocyanins, confirmed by the lowest measured PPC.

**KEYWORDS:** *Aronia melanocarpa*, polyphenols, anthocyanins, extraction, solvent, degradation index

### CHEMICAL COMPOSITION OF VOLATILE COMPOUNDS OF EIGHT GERANIUM L. SPECIES FROM VLASINA PLATEAU

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The Vlasina plateau shows significant floristic diversity counting 956 species of which 91 may be categorized as endangered in the flora of Serbia. The genus *Geranium* is present in the flora of this region with few species and they are the subject of our research. The chemical composition of volatile compounds of aerial and underground parts of eight *Geranium* L. species (*G. macrorrhizum* L., *G. phaeum* L., *G. sanguineum* L., *G. robertianum* L., *G. palustre* L., *G. pyrenaicum* Burm. f., *G. columbinum* L. and *G. lucidum* L.) collected in flowering stage from Vlasina plateau (Serbia) was examined. The samples of volatile fraction were obtained by steam distillation and analyzed using GC-FID and GC-MS. The cluster analysis revealed the separation of volatile compounds of *Geranium* species to two clusters. The aerial and underground parts volatiles of *G. macrorrhizum* were separated from the other species, based on the high sesquiterpenes content (92.3% and 94.6%), mostly germacrene (73.3%) in the aerial and  $\delta$ -guaiene (76.8%) in the underground parts. The volatile fractions of other species were mainly composed of sesquiterpenes (10.8-61.8%), diterpenes (12.9-43.0%) and fatty acids and their derivatives (6.6-21.6%). The exception was the fraction of the underground parts of *G. phaeum* which was predominantly made by fatty acids and their derivatives (76.6%). In the volatile fraction of *G. palustre* aerial parts  $\beta$ -selinene (18.6%) and (*E*)-caryophyllene (15.7%) prevailed. Hexadecanoic acid (15.4%), germacrene D (15.4%), (*E*)-caryophyllene (10.6%), phytol (10.4%) and caryophyllene oxide (10.2%) were the most abundant compounds in volatile fraction of *G. pyrenaicum* aerial parts, while phytol (13.2%) and (*E*)-caryophyllene (9.5%) in the volatile fraction of *G. lucidum* aerial parts. Hexadecanoic acid and phytol were dominant compounds in the volatile fraction of aerial parts of *G. sanguineum* (21.1% and 17.8%), *G. robertianum* (12.5% and 19.3%), *G. phaeum* (7.3% and 41.4%) and *G. columbinum* (14.5% and 29.5%). Volatiles of aerial parts of *G. robertianum* were also characterized by (*E*)-caryophyllene (8.0%) and caryophyllene oxide (7.4%), while the volatiles of *G. sanguineum* contained  $\beta$ -bisabolene (5.2%). The volatiles of *G. palustre* aerial parts have been studied for the first time. The chemical characterization of volatile compounds of *Geranium* species may possibly have chemotaxonomic importance and is significant for further research of Vlasina plateau native flora.

**KEYWORDS:** *Geranium*, Vlasina plateau, volatile fraction, GC-FID, GC-MS

### CHEMICAL PROFILE OF BLACK CUMIN SEED OILS AND EXTRATCS

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The chemical composition of essential oil (NS-HD), fixed oil (NS-SH), methanol (NS-SM) and water (NS-RW) extracts of black cumin (*Nigella sativa* L.) seed were evaluated. The volatile constituents of essential and fixed oils, as well as fatty acid profile in fixed oil were determined via gas chromatography-mass spectrometry (GC/MS) technique. Phenolic acids and flavonoids were determined in methanol and water extracts of *N. sativa* seed by UHPLC-UV/Vis method. Essential oil showed the presence of eleven compounds representing 80.92% of the total amount. The major component of the essential oil was *p*-cimene (46.18%), followed by  $\alpha$ -thujene (8.68%), longifolene (7.32%), and thymoquinone (6.77%). Analysis of the fixed oil resulted in the identification of nineteen compounds representing 97.13% of the total amount; and linoleic acid (45.29%), oleic acid (23.33%), *p*-cimene (9.26%) and thymoquinone (7.33%) were found as the main components. Nine fatty acids were identified in the fixed oil. The extract was consisted of four saturated fatty acids (24.43%) and five unsaturated fatty acids (75.57%). Linoleic acid (24.58%), oleic (14.11%) and eicoseic acid (12.00%) were the major components. Eight phenolic acids (gallic, 4-hydroxybenzoic, chlorogenic, vanillic, caffeic, ferulic, sinapinic, and rosmarinic acid) were found in water and methanol extracts. Vanillic acid was the main component of both extracts (5.79  $\mu$ g/g for NS-SM, and 4.63  $\mu$ g/g for NS-RW). Regarding the flavonoids profile, quercetin was identified only in NS-SM extract.

**KEYWORDS:** *Nigella sativa* L., volatile compounds, fatty acids, phenolic acids, flavonoids

Poster presentation 34 06 44

### CHEMICAL COMPOSITION AND ANTIOXIDANT ACTIVITY OF *GERANIUM MACRORRHIZUM* L. ESSENTIAL OIL IN RELATION TO PLOIDY LEVEL AND ENVIRONMENTAL CONDITIONS

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*Geranium macrorrhizum* L. is an aromatic plant native to the Southern Alps and the Balkan Peninsula. In Europe it is increasingly cultivated on account of its ornamental flowers, as well as for its use in traditional herbal medicine and aromatherapy. This aromatic plant possesses a broad spectrum of antimicrobial, hypotensive, spasmolytic, astringent, cardiotonic, antioxidant, capillary, and sedative activities. Essential oil of *G. macrorrhizum* is highly valued in perfumery due to its excellent fixative properties, and it is also used as a food-flavoring agent. In this presentation we report the essential oil composition, total phenolics and antioxidant activity of several samples of *G. macrorrhizum* that were collected from natural habitats in Croatia (Mt Biokovo) and Greece (Mt Smolikas and Mt Olimbos). These were then transferred to the greenhouse or botanical gardens in separate locations in Bosnia and Herzegovina (Sarajevo), France (Orsay) and Denmark (Copenhagen). Since the plant samples had different cytotypes, we examined a possible correlation between ploidy level, phenolic content, and antioxidant activity. We found that the total phenolic content of hydrosols of *G. macrorrhizum* is related to plant ploidy level, e.g., hexaploid samples contained almost twice the amount of phenolic compounds than hydrosols of diploids and tetraploids. Radical scavenging activity of the same extracts revealed similar correlations. However, the essential oil composition clearly depends on environmental factors. The major compounds in almost all essential oils of transplanted plants were oxygenated sesquiterpenes with germacrene as the most abundant (34.7-62.9%). The essential oil of the plants collected from natural habitats was rich in long-chain alkanes (27.0-40.0%). Antioxidant activity of the essential oils was notably weaker than for corresponding hydrosols; this is related to the low concentrations of terpenoids that are able to scavenge stable radicals.

**KEYWORDS:** *Geranium macrorrhizum* L., ploidy level, essential oil composition, total phenolic content, antioxidant activity

Poster presentation 35 06 50

### VOLATILE COMPOUNDS OF NEWLY DISCOVERED SPECIES *CENTAUREA ZLATIBORENSIS* (ASTERACEAE)

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In this paper, the composition of essential oil from fresh capitula of *Centaurea zlatiborensis* Zlatković, Novaković & Janačković, sp. nova (*Centaurea* sect. *Acrocentron*, Asteraceae) an endemic species from Mt. Zlatibor (Serbia), was isolated using Likens-Nickerson type apparatus and analyzed by gas chromatography coupled with flame ionization detector (GC-FID) and by gas chromatography coupled to mass spectrometry (GC-MS). Fifty-four compounds were determined representing 97.8% of the total oil. The overall composition of essential oil of *C. zlatiborensis* is characterized by a high percentage of sesquiterpenes (81.9%) and a low percentage of monoterpenes (1.3%). The dominant components were two sesquiterpene hydrocarbons: (E)-caryophyllene (27.88%), germacrene D (17.05%), and one oxygenated sesquiterpene - caryophyllene oxide (9.41%). Other compounds (aliphatic hydrocarbons, aliphatic aldehydes and alcohols, aliphatic acids and their esters and aldehydes, aromatic esters and aliphatic acids, alkyl aromatic alcohols, aryl esters of aromatic acids) represent 8.7%. According to results of our previous investigations and literature data, essential oils of the most *Centaurea* species from the section *Acrocentron* were characterized by the presence of (E)-caryophyllene and germacrene D as dominant compounds. Future work will be directed toward the chemotaxonomic significance of volatile compounds of other *Centaurea* species from the section *Acrocentron*, as well as from related sections.

**KEYWORDS:** *Centaurea zlatiborensis*, Asteraceae, essential oil

Poster presentation 36 06 51

### PRELIMINARY ANALYSIS OF SPECIALISED METABOLITES OF *CYTISUS JANKAE* FLOWERING AERIAL PARTS

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*Cytisus jankae* Velen. (Fabaceae) is a dwarfish shrub (10–25 cm height) with 3-foliolate leaves and capitate inflorescence with tubular calyces. The whole plant is densely covered with silky hairs. Its range includes central and east part of the Balkan Peninsula and Romania. It inhabits dry rocky places on limestone or ultramafic soils. The objective of this study was to establish the phytochemical profile of flowering aerial parts of this plant. The plant material was collected in eastern Serbia (Jelašnička Klisura gorge). In the air-dried herb, by the direct gravimetric method, the total alkaloids content (0.32%) was determined, and using colorimetric assays the total polyphenols (43.8%), tannins (1.13%), and the total flavonoids (1.31%) were quantified. Additionally, the powdered dried plant material was successively extracted with dichloromethane (maceration) and methanol (bimaceration) at room temperature. After the solvents were evaporated under reduced pressure, dried extracts were subjected to the further analysis. The total alkaloids content in both the dichloromethane (7.01%) and methanol (3.47%) extracts, and the contents of total polyphenols (13.75%), tannins (2.45%) and flavonoids (4.65%) in the methanol extract, were determined by aforementioned tests. Applying the LC-MS method, in the methanol extract flavonoids quercetin 3-O-rutinoside, luteolin, apigenin and genistein were identified. GC-MS analysis resulted in the identification of quinolizidine alkaloids sparteine, 17-oxosparteine and lupanine (2-oxosparteine) in both the methanol and dichloromethane extracts, as well as of one triterpene, β-amyrin, in the dichloromethane extract. This is the first report on specialised metabolites from flowering aerial parts of *C. jankae*.

**KEYWORDS:** *Cytisus jankae*, flowering aerial parts, flavonoids, LC-MS, alkaloids, GC-MS

Poster presentation 37 06 62

### ANTHOCYANIN PROFILE OF CORNELIAN CHERRY (*CORNUS MAS* L.) FRUITS

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Cornelian cherry (*Cornus mas* L.) is deciduous small tree native grown in central and southeastern Europe and southwestern Asia. Its fruits were traditionally used mainly for gastrointestinal disorders and diabetes treatment. Scientific data on compound profile of cornelian cherry is still insufficient, especially for Serbian genotypes. Anthocyanins are phenolic compounds responsible for purple-red colour of fruits and vegetables. They have many beneficial effects on human health, especially as free radical scavengers, vasoprotective, anticarcinogenic and antidiabetic agents. The objective of this research was to determine anthocyanin content of cornelian cherry fruits from Serbia. Seven cornelian cherry genotypes grown in Vojvodina (northern Serbia) were assessed: Apatinski, Bačka, Krajišnik, Rus, Elegantni, Semen, S2. Fresh fruits collected in September 2016. were lyophilized and grinded prior to extraction. Powdered samples were ultrasonicated in 50% ethanol (1:10, w/v), evaporated and redissolved in water. Samples were analysed by RP-HPLC-PDA method. Individual and total anthocyanins were detected at 520 nm and quantified as mg cyanidin 3-glucoside equivalents per gram of lyophilized fruits (mg/g LF). S2 was distinguished as the genotype most abundant in anthocyanin content (10.81 mg/g LF). The second abundant one, Semen genotype, possessed three times less anthocyanins (3.68 mg/g LF), while other genotypes contained from 0.73 mg/g LF (Rus) to 2.44 mg/g LF (Elegantni). All fruit extracts showed exactly the same anthocyanin profile, varying just in quantitative content. Only two anthocyanins (cyanidin 3-O-galactoside and pelargonidin 3-O-galactoside) contributed over 95% of total anthocyanin content among all genotypes. In elegantni genotype only, content of these two glycosides was equal (approx. 47% of each). Cyanidin 3-O-galactoside dominated in genotype S2 (71.7%), while pelargonidin 3-O-galactoside made 81.6% of total anthocyanins in Apatinski genotype. Further studies on expanded number of cornelian cherry genotypes, could provide detailed data on possible usage of anthocyanin profile in chemotaxonomic purposes, besides from their health promoting potential.

**KEYWORDS:** cornelian cherry, cyanidin 3-O-galactoside, pelargonidin 3-O-galactoside



Poster presentation 38 06 61

**VOLATILE COMPOUNDS OF THE COMMON SPEEDWELL (*VERONICA ARVENSIS* L.)**Marija Nazlić<sup>1\*</sup>, Dario Kremer<sup>2</sup>, Mirko Ruščić<sup>1</sup>,  
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Genus *Veronica* L. is divided into 13 subgenera according to morphological traits and it belongs to the Plantaginaceae family. There are 40 species of *Veronica* in Croatia. They are cosmopolitan and ecologically diverse species spread on a variety of habitats from aquatic, marshy and forest to rock, rock cracks, fields and ruderal habitats. Investigated *Veronica arvensis* L. belongs to terophytes. Literature review showed that the most investigated secondary metabolites for *Veronica arvensis* and generally for *Veronica* genus include iridoid glucosides, phenylethanoids and flavonoid glycosides, so this is the first study of the chemical composition of the essential oil from this species. Plant material was collected from Velebit (Croatia), in the spring (June) 2013. and was air dried. Water distilled essential oil from aerial parts of investigated plant have been analyzed by GC (gas chromatography) and GC/MS (gas chromatography – mass spectrometry) using VF-5ms capillary column and the first peaks appear after 30 minutes. The total yield of oil was 0.1%, based on dry weight of samples. Nine compounds were determined, representing 78,4% of the total oil. Results of the GC/MS showed that the most abundant compound is phytone represented with 38,1% followed by hexadecanoic acid (26,3%) and hexahydrofarnesyl acetone (4,4%). In our previous research in the oil of *V. spicata* phytol was the most abundant compound represented with 21.3%. In our review, we found one more species, *Veronica thymoides* subsp. *pseudocinerea*, that was investigated with the same methodology (GC-MS). Compound peaks for *Veronica thymoides* also appeared after 30 minutes of the GC-MS analysis and the most abundant constituent was hexatriacontene (21%). The present study gives additional knowledge about volatile compounds of the genus *Veronica*.

**KEYWORDS:** *Veronica arvensis*, Plantaginaceae, volatile compounds, secondary metabolites, phytone

Poster presentation 39 06 66

**GC/MS BASED METABOLITE PROFILING OF BULGARIAN *MICROMERIA* SPECIES**Milena Nikolova<sup>1</sup>, Ina Aneva<sup>1\*</sup>, Irena Mincheva<sup>1</sup>,  
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The species of genus *Micromeria* Bentham provoke interest from phytochemical point of view worldwide due to their variable biological activities and potential for use in the pharmaceutical industry. In Bulgaria genus *Micromeria* is represented by 4 species: *M. cristata* (Hampe) Griseb., *M. dalmatica* Benth., *M. frivaldszkyana* (Degen) Velen. and *M. juliana* (L.) Benth. ex Rchb. Ten populations - one of *M. frivaldszkyana*, two of *M. juliana*, three of *M. cristata*, four of *M. dalmatica* were analyzed for metabolites by GC/MS. Alkanes, fatty alcohols, fatty acids, triterpenes, phenolic acids, organic acids, carbohydrates and phytosterols were identified. Seventeen saturated and unsaturated fatty acids were identified. Palmitate (C16:0), linolenic (C18:3) and linoleic (C18:2) being the main acids. In all species  $\beta$ -sitosterol was present in significant amounts.  $\beta$ -Amyrin and triterpene derivatives were present in a large amount of studied extracts too. Sucrose was the main carbohydrate in all samples. 4(p)-hydroxybenzoic acid and vanilic acids were common phenolic acids for all studied species. Caffeic and p-hydroxycinnamic acids were present in the largest quantities in *M. dalmatica* populations. Organic acids as well as chlorogenic acid are characteristic mainly of *M. dalmatica* and *M. frivaldszkyana*. The results showed that *Micromeria* species and especially *M. dalmatica* are rich in bioactive compounds such as triperpens (amyrin,  $\beta$ -sitosterol), omega-3 fatty acids (linolenic acid), phenolic and organic acids that determines their value as food and medicinal plants.

**ACKNOWLEDGMENTS:** Program for career development of young scientists, BAS Grant № DFNP-67\_A1**KEYWORDS:** medicinal plants, GC/MS, metabolites

Poster presentation 40 06 74

**PHYTOCHEMICAL ANALYSIS OF *MENTHA PULEGIUM* L. (LAMIACEAE) POPULATIONS FROM PANNONIAN PLAIN, THE CARPATHIAN MT AND BALKAN PENINSULA**Bojana Bokić<sup>1\*</sup>, Nebojša Kladar<sup>2</sup>, Milica Rat<sup>1</sup>,  
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*Mentha pulegium* L. is a perennial, aromatic and culinary herb, very variable in habit, leaf-shape and hair type. It occurs mostly on ruderal and riparian habitats in southern, western and central Europe, northern Africa and Middle East as a native species. Phenolics play an important role for plants. They act as signaling molecules, defensive compounds or anti-stress agents, and generally are included in growth and resistance of plants. However, they display a great heterogeneity and their presence and amounts are subject of variations among plant organs, ages, seasons and environmental conditions. Also, phenolics are considered as valuable phytochemical markers. Therefore, our objective was chemical characterization of *M. pulegium* wild populations in order to determine their diversity in the aspect of phenolic compounds, as well as to detect those that can (not) be used for species and interspecies differentiation. Aerial parts of plant populations at 21 localities were collected during 2013-2016 and air dried. Plant extracts were made using a method of maceration with methanol for 24 h at room temperature, filtered and evaporated to dryness. The amounts of total phenolics and total flavonoids were quantified spectrophotometrically and expressed as mg of gallic acid equivalents (GAE) per g of dry extract (d. e.) and mg of quercetin equivalents (QE) per g of d. e., respectively. Detailed chemical characterization of the examined extracts included quantification of apigenin, naringenin, epicatechin, quercetin, rutin, as well as ferulic, chlorogenic, caffeic, coumaric, rosmarinic, sinapic and vanillic acids by high-performance liquid chromatography coupled with diode array detection (HPLC-DAD). Statistical analyses were performed in PAST ver. 3.20 and STATISTICA ver. 13.3. Methanol extracts obtained from *M. pulegium* collected at different localities contain high amounts of total phenolic and flavonoid compounds, with significant variations among populations. The amount of total phenolics ranged from 41.88 to 290.74 mg of GAE/g d. e., while the content of total flavonoids ranged from 9.12 to 138.90 mg of QE/g d. e. The quantified amounts of rutin, apigenin, quercetin and coumaric acid indicate several groups, which have similar chemical profiles within *M. pulegium*, and this can be potentially important for its ecological differentiation.

**KEYWORDS:** HPLC, phenolics, flavonoids, Serbia, Montenegro, European pennyroyal, Labiatae

Poster presentation 41 06 42

**CYCLAMEN *HEDERIFOLIUM* AIT. METAL COMPOSITION**Ksenija Kojičić<sup>1\*</sup>, Snežana Cupara<sup>1</sup>, Snežana Branković<sup>2</sup>,  
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The species *Cyclamen hederifolium* Ait. belongs to the family Primulaceae, which comprehends 23 species of cyclamen, naturally distributed in the southern parts of Europe, Western Asia and parts of North Africa. It is considered as a poisonous plant, though extracts of *C. hederifolium* tubers have been used sporadically in Serbian traditional medicine as purgative and anti-tumor agents, in the treatment of rheumatism, migraine and skin conditions. Biological activities of *C. hederifolium* have been attributed to the triterpenic saponosides. The content of metals has not been documented thoroughly for this species. The aim of this paper was to evaluate content of metals (Mn, Ca, Mg, Fe, Zn, K, Cu, Cr, Ni, Pb, Cd) in the *C. hederifolium* growing on Stara Planina in Eastern Serbia and to determine possible phytoaccumulation of metals in *C. hederifolium* tuber in the comparison to soil and plant extracts (aqueous and ethanol). The flame atomic absorption spectrophotometer (FAAS) was used for the determination of metals. In order to determine the total metal content in soil, the samples were prepared by digestion with nitric acid and hydrogen peroxide. The values obtained in the range of  $\pm$  5% of the certified values. The values for Cr, Ni, Pb and Cd were below the detection limit. The highest values of Ca, Fe and Mn were found in soil, Mg and Zn in aqueous extract, and K in tuber. The aqueous extract had a higher content of all observed metals than ethanol extract, except Cu. Compared to soil, ethanol extract contained more Mg and K and aqueous extract more Mg, Zn and K. The concentrations of investigated metals found in tuber were in descending order: Ca>Mg>K>Fe>Mn>Zn>Cu. It has been shown that tuber contains more Ca, K and Cu than aqueous and ethanol extracts, as well as Mg and Mn than ethanol extract. Higher concentrations of Mg and K found in tuber suggest potential phytoaccumulation.

**KEYWORDS:** *Cyclamen hederifolium* Ait., tuber, metals, phytoaccumulation

Poster presentation 42 06 13

### TEPALS OF NATIVE *CROCUS* TAXA AS A PROMISING ANTIOXIDANT BIOMATERIAL RICH IN FLAVONOID KAEMPFEROL

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Most of the pharmacological activities of cultivated *Crocus sativus* have mainly been related to its stigmas (spice saffron) and their major bioactive compounds - carotenoids. Tepals, which are dominant part of *C. sativus* flowers in terms of mass (93% of the flower mass), are still considered as a waste material. In this work we compared, using high-performance liquid chromatography method, qualitative and quantitative composition of flavonoids in tepals, stigmas and stamens of native *Crocus* taxa (*Crocus malyi*, *Crocus vernus* ssp. *albiflorus*, and *Crocus vernus* ssp. *vernus*) water extracts, spectrophotometrically measured antioxidant activities of tepals extracts, and statistically correlated obtained data. The results revealed that *Crocus* tepals contained similar or higher concentration of flavonoid-glycosides than stigmas and stamens. Out of the tested tepal extracts, *C. vernus* ssp. *vernus* from the Botanical Garden Zagreb showed the best biological potential: the highest amount of identified flavonoids (5054.6 mg of identified flavonoid-glycosides/kg dw) and showed the highest antioxidant activity (ABTS - 82%, FRAP - 99.2% , and DPPH - 62.7% of Trolox antioxidant activity). In this taxon we also recorded significantly higher concentration of kaempferol-rutinoside (3581 mg/kg dw) than in other taxa. This flavonoid showed very strong or strong correlation with antioxidant assays results. Therefore, we presume that kaempferol-rutinoside is one of the main antioxidant phenolic component in *Crocus* tepals. High flavonoid content and strong antioxidant activity add value to the, so far, neglected native *C. vernus* ssp. *vernus* taxon. To the best of our knowledge, this is the first study on native Croatian *Crocus* taxa flavonoid content, and their bioactivity at all. The results contribute to the phytochemical description and dietary potential of native *Crocus* taxa.

**KEYWORDS:** dietary potential, HPLC, phenolics, phytochemical analysis, saffron

Poster presentation 43 06 28

### ANTIOXIDANT POTENTIAL AND POLYPHENOLIC CONTENT OF THREE MEDICINAL WILD-GROWING SPECIES FROM KOPAONIK MOUNTAIN

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Wild-growing plants are good potential sources of natural substances useful for preventing diseases related to oxidative stress. The food industry is becoming increasingly interested in plants, because of their anti-inflammatory properties and antioxidant activities. In the present investigation, we described the antioxidant potential of acetone extracts of three aromatic species grown on Kopaonik mountain, *Ajuga pyramidalis* L., *Thymus praecox* Opiz and *Vaccinium myrtillus* L. by two methods: 2,2-azinobis(3-ethyl-benzothiazoline-6-sulfonic acid) (ABTS) scavenging and 2,2-diphenyl-1-picryl-hydrazyl (DPPH) radical scavenging assay and their correlations with total phenolics, tannins, flavonoids and proanthocyanidins contents. The total phenolic and tannins content was determined according to the Folin-Ciocalteu method. The total flavonoids were estimated according to the method described by Markham (1989) and proanthocyanidins according to Sun et al. (1998). For ABTS assay, the procedure followed the method of Re et al. (1999) with some modifications. The DPPH assay was done according to the method of Lee et al. (1998). *T. praecox* extract had the almost twice higher phenolics and tannins content compared to *V. myrtillus*, which had twice higher total phenolics and tannins content than *A. pyramidalis*. On the other hand, *A. pyramidalis* had the highest amount of proanthocyanidins, while the content of total flavonoids was the highest in *V. myrtillus*. The highest scavenging activity showed *T. praecox* extract (DPPH: 69,67±0,46 mg trolox/gdw; ABTS: 178,11±0,96 mg trolox/gdw), and the lowest activity showed *A. pyramidalis* extract. Antioxidant capacity was positively correlated with total phenolics, tannins and flavonoids content. From the obtained results it can be concluded that *T. praecox* extract had the greatest antioxidant potential.

**KEYWORDS:** antioxidant potential, medicinal plants, polyphenolic content

Poster presentation 44 06 03

### A COMPARATIVE STUDY ON ANTIOXIDANT PROPERTIES OF METHANOL AND WATER EXTRACTS OF *ORIGANUM ONITES* L. (LAMIACEAE) FROM TURKEY

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*Origanum onites* L. (Lamiaceae), as called Turkish Oregano, is a perennial plant of the east Mediterranean area. It is consumed intensively as a spice especially by the people in the western regions of Anatolia. This work aimed to evaluate the antioxidant activity of methanol and water extracts of wild-grown *O. onites* collected from Fethiye region (Muğla, Turkey), as well as to measure the content of total phenolic compounds of these extracts. The extracts were obtained by ultrasonication method in this study. Total phenolic content present in the extract was also determined by Folin-Ciocalteu assay. Antioxidant activities were investigated by using different assays, including free radical scavenging assays (ABTS and DPPH) reducing power (CUPRAC and FRAP), phosphomolybdenum and metal chelating assays. The total phenolic contents were found to be 86.40 and 111.41 mgGAE/g extract in methanol and water extracts, respectively. Free radical scavenging ability of water extract (432.99 mgTE/g for ABTS and 243.37 mgTE/g for DPPH) was higher than methanol extract (353.68 mgTE/g for ABTS and 193.61 mgTE/g for DPPH). Likewise, the water extract had greater reducing potential in CUPRAC and FRAP assays, as compared to methanol extract. The results of phosphomolybdenum were found to be 2.29 mmolTE/g for methanol and 2.68 mmolTE/g for water. Taken together, the tested water extract had greater antioxidant potential when compared to methanol extract. Based on our findings, *O. onites* could be regarded as a source natural bioactive agent for developing novel functional ingredients.

**KEYWORDS:** free radical scavenging, *Origanum onites*, total phenolic, natural agents

Poster presentation 45 06 05

### ANTIOXIDANT AND ENZYME INHIBITORY FUNCTIONS OF THREE SELECTED *FERULA HALOPHILA* (APIACEAE) PEŞMEN EXTRACTS

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Currently, many plant bioactive compounds are considered as excellent alternative candidates to synthetic antioxidants or antimicrobials food additives possessing none side effects in comparison with synthetic additives. In this sense, medicinal plants are seen as an important and feasible alternative, as being used for thousands of years for the treatment and prevention of several health disorders. The genus *Ferula* (Apiaceae) has great potential as a source of traditional drugs in several countries. With this fact, we aimed to determine antioxidant and enzyme inhibitory functions of three solvent extracts (acetone, chloroform and methanol) of *F. halophila*, which is endemic to Turkey. Six complementary assays (DPPH, ABTS, FRAP, CUPRAC, phosphomolybdenum and metal chelating) were performed to obtain full antioxidant picture for the tested extracts. Cholinesterase (acetylcholinesterase (AChE) and butyrylcholinesterase (BChE)), tyrosinase,  $\alpha$ -amylase and  $\alpha$ -glucosidase were selected as target enzymes. The acetone and methanol extracts had greater antioxidant potential as compared to chloroform extract. Also, these extracts contained considerable levels of phenolics (55.22 mgGAE/g extract for acetone; 48.66 mgGAE/g extract for methanol). However, the chloroform extract was more active on both AChE and BChE than the acetone and methanol extracts. The best tyrosinase inhibitory effect was obtained by the methanol extract. Overall, *F. halophila* could be regarded as a valuable source of novel phyto-pharmaceuticals for designing functional formulations, including food additives or drugs.

**KEYWORDS:** *Ferula halophila*, tyrosinase, cholinesterase, antioxidant

Poster presentation 46 06 21

### ANTIOXIDANT ACTIVITY OF IMMORTELLE EXTRACTS (*HELICHRYSUM ITALICUM* (ROTH) G. DON FIL.)

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*Helichrysum italicum* (Roth) G. Don fil. (family Asteraceae), also called immortelle, is one of the most studied species of the genus *Helichrysum*. It is widely distributed in the Mediterranean region, growing on sunny, rocky slopes and around sandy areas. Traditionally, immortelle is being used for the treatment of scars and all kind of cuts. The drug is widely used as a liver stimulant and diuretic, but also as an antioxidant, antiallergenic, antimicrobial, antiviral, choleric, and antihypertensive agent. Therefore, the purpose of this study was to evaluate antioxidant activity of immortelle extracts along with total phenolic and flavonoid contents obtained from different concentration of ethanol. The extraction was performed with water and ethanol during different periods of time (10, 30, 60 min and 24h) and the degree of fragmentation was 0.3 and 2 mm. Antioxidant capacity was investigated by spectrophotometric measurement of total phenolic and flavonoid content and inhibitory activity of DPPH radical. The obtained results demonstrated that analyzed extracts exerted significant antioxidant activity. Total phenol content ranged from 14,83 – 18,61 mg GAE/gDE, while content of total flavonoids ranged from 1,57 to 22,23 mg QE/gDE. Generally, all extracts showed good antioxidant activity with an IC<sub>50</sub> value in the range from 38,21 to 61,82 µg/ml. In conclusion, it was found that increased time of extraction, concentration of ethanol and degree of comminution of the drug increase the quality of the extracts in terms of the content of phenolic components and antioxidant effects.

**KEYWORDS:** *Helichrysum italicum*, antioxidant activity, phenolic, flavonoid DPPH

Poster presentation 47 06 34

### REACTIVE OXYGEN SPECIES AND ANTIOXIDATIVE DEFENSE MECHANISM IN WATER-STRESSED SOYBEAN (*GLYCINE MAX L.*)

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Environmental stresses in addition to their direct effects on plants growth and development induce oxidative stresses on plants. These secondary stresses cause cell wall lipids and cell organelles proteins degradations. In response to overflow of reactive oxygen species (ROS) enzymatic and non-enzymatic antioxidants productions induces in plants. Therefore, the aim of this research was to evaluate the effect of soil water shortage on soybean antioxidants induction. This research was conducted on spring and summer 2012 in Sari Agricultural Sciences and Natural Resources University (SANRU), Sari, Iran. The JK variety of soybean (*Glycine max L.*), widely cultivated in the region, was selected for this purpose. A completely randomized design with 4 water level treatments in a pot experiment was employed. After 4-leaf stage soybean seedlings were randomly assigned to 4 water treatments of control, with 100 (well-watered, W0), 60 (mild stressed, W1), 40 (intermediate stressed, W2) and 20 percent (severe water stressed, W3) of soil field capacity (FC). Plants re-watered to 100 percent FC upon reaching to the assigned water level. During R<sub>4</sub> and R<sub>5</sub> reproductive stages leaf samples were collected to measure the content of leaf enzymes and proteins spectrophotometrically. Data were analyzed using SAS software and mean comparisons were done using Tokay post-hoc test. Findings of this research showed that catalase enzyme activity increased up to 40% FC, then declined by further reduction in soil available water. Also, the trend of variations in peroxidase activity, malondialdehyde (MDA) values and leaf total nitrogen contents increased by reduction in soil available water, while the trend was reverse for leaf soluble proteins. The maximum activity of catalase was achieved by moderate water stress, while the maximum activity of peroxidase enzyme, the highest MDA values and total leaf nitrogen contents, as well as the lowest leaf soluble proteins were achieved by severe water stress treatment.

**KEYWORDS:** water stress, antioxidant, peroxidase, catalase, soybean, ROS

Poster presentation 48 06 40

### EVALUATION OF ENZYME INHIBITORY EFFECTS AND ANTIOXIDANT POTENTIAL OF *CENTAUREA DEFLEXA* WAGENITZ AND *CENTAUREA KOTSCHYI* (BOISS. & HELDR.) HAYEK VAR. *KOTSCHYI*, GROWING IN TURKEY

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*Centaurea L.* with about 500 species in the world, is one of the most widespread members of the Asteraceae family plants. In Turkey, the genus is represented by 194 species, 118 of them are endemic. Some *Centaurea* species have been used as herbal remedies in Anatolian folk medicine to treat fever, common cold, diabetes, hemoroid, abscesses and peptid ulcer. Many studies on *Centaurea* species have identified the presence of seconder metabolites, such as esquiterpene lactones, flavonoids, lignans, alkaloids, phenolic compounds and steroids. There is no extensive evaluation of biological activities on *Centaurea deflexa* Wagenitz and *C. kotschyi* (Boiss. & Heldr.) Hayek var. *kotschyi*. The aim of our study was to assess *in vitro* antioxidant and tyrosinase inhibition activities as well as determine their content of total phenolic compounds (TPC) and flavonoids (TF) of aerial parts of *C. deflexa* and *C. kotschyi* var. *kotschyi*. The methanol and water extracts of the plants were analysed for the antioxidant activities using free radical scavenging methods, including DPPH and ABTS, and their content of TPC and TF were determined spectrophotometrically. According to our results, the content of total phenolic compounds and flavonoids ranged between 18.98 ± 0.77 and 43.86 ± 4.53 mg gallic acid equivalent per gram of extract, and 31.68 ± 7.47 and 52.43 ± 2.85 mg quercetin equivalent per gram of extract, respectively. As comparing with *C. deflexa*, it was found that *C. kotschyi* var. *kotschyi* has higher amount of TPC and TF. Among the samples, the highest DPPH and ABTS free radical scavenging activities were displayed in the methanol extract of *C. kotschyi* var. *kotschyi* with IC<sub>50</sub> values of 1323.04 ± 0.75 µg/mL and 85.80 ± 6.31 µg/mL, respectively. In addition, the methanol extract of *C. kotschyi* var. *kotschyi* showed the highest tyrosinase inhibition with IC<sub>50</sub> value 1.031 ± 1.22 mg/mL. As a conclusion, *C. kotschyi* var. *kotschyi* can be regarded as a potential source of antioxidant compounds and tyrosinase inhibitors to develop new pharmaceutical medicines. Further studies need to be conducted on identification of bioactive constituents for these biological activities.

**ACKNOWLEDGEMENTS:** This study was supported by Selcuk University Research Foundation [18401110].

**KEYWORDS:** tyrosinase inhibition, *Centaurea deflexa*, *Centaurea kotschyi* var. *kotschyi*, radical scavenging activity, total phenolic and flavonoids

Poster presentation 49 06 73

### CHANGES IN ANTIOXIDANT ENZYME ACTIVITY IN RESPONSE TO COLD-INDUCED OXIDATIVE STRESS OF *OCIMUM BASILICUM L. VAR. PURPURASCENS* BENTH.

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Low temperature is one of the primary abiotic stresses that limit plant development and affect crop productivity. Oxidative stress is one of the deleterious aspects of cold stress, which facilitates the formation of reactive oxygen species (ROS) and subsequently damages to cell structures. Under cold stress, plants have some effective intracellular mechanisms to alleviate oxidative damages, including enzymatic antioxidant enzymes that were shown to be activate dramatically in cold-stressed plants. *Ocimum basilicum L. var. purpurascens* Benth., commonly known as purple basil, is one of the most widely used basil cultivar with high economic value throughout the world. However, its growth and development are often influenced by temperature. The main object of this work was evaluation of antioxidant enzyme activities in leaves and roots of cold-stressed *O. basilicum* var. *purpurascens* (cv. Dark Opal). Low temperature treatment (10 °C and 4 °C) were conducted on five weeks old seedlings from *O. basilicum* var. *purpurascens* (cv. Dark Opal). Antioxidant enzyme activities were evaluated spectrophotometrically in leaves and roots and compared with control plants (25 °C). The responses of antioxidant enzymes were variable regarding the treatments as well as investigated seedling parts. Superoxide-dismutase (SOD) was induced only in roots under 4 °C. Activity of catalase (CAT) increased in leaves under both treatments were in roots was induced only under severe temperature stress. Ascorbate-peroxidase (A-POX) was the most cold-sensitive enzyme examined, with decreased activities under 10 °C and 4 °C in leaves and roots, respectively. By contrast, pyrogallol-peroxidase (P-POX) and guaiacol-peroxidase (G-POX) activity increased several times in cold-stressed purple basil roots. The changes in nonspecific antioxidant biomarkers observed in this study could be highly valuable for studying the mechanism underlying cold-induced tolerance in *O. basilicum* var. *purpurascens*.

**KEYWORDS:** purple basil, low temperature, stress, antioxidant enzyme

### ANTIMICROBIAL ACTIVITY OF *ORIGANUM VULGARE* L. ESSENTIAL OIL

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*Origanum vulgare* L. is perennial herbaceous well-known medicinal plant belonging to the family Lamiaceae that has been used since ancient times as an additive in foods and cosmetic preparations. Since oregano essential oils are widely used in the treatment of respiratory, urinary and cutaneous infections in traditional medicine the aim of this study was to evaluate the antimicrobial activity against the wide range of pathogenic microorganisms. In order to evaluate antimicrobial activity of essential oregano oil disc-diffusion method was applied. A disc containing ampicillin, streptomycin, and nystatin were used as positive control. Various concentrations of the essential oil (100%, 75%, 50%, 25%) were used for antimicrobial susceptibility testing. The screening of antibacterial and antifungal activity was evaluated against four Gram positive bacteria: *Staphylococcus aureus* ATCC 25923, methicillin-resistant *Staphylococcus aureus* (MRSA) ATCC 33591, *Bacillus subtilis* ATCC 6633, *Enterococcus faecalis* ATCC 29212 and five Gram-negative bacteria: *Salmonella abony* ATCC 6017, *Salmonella enterica* serovar Enteritidis ATCC 31194, *Pseudomonas aeruginosa* ATCC 9027, *Escherichia coli* ATCC 25922, extended-spectrum  $\beta$ -lactamase (ESBL) producing *Escherichia coli* ATCC 35218 and fungi *Candida albicans* ATCC 1023. In our study, all tested microorganisms were extremely inhibited by essential oregano oil. The highest activity of oregano essential oil is found in the case of *S. aureus*, MRSA, *Bacillus subtilis* and *Candida albicans*. On the other hand, the smallest inhibition zone was recorded for *Salmonella enterica* serovar Enteritidis. The results of antimicrobial assay revealed that tested Gram positive bacteria were more susceptible to investigated essential oil than Gram negative bacteria. In conclusion, oregano essential oil had strong antimicrobial effects on a variety of human pathogenic microorganisms, including MRSA and ESBL producing *Escherichia coli*.

**KEYWORDS:** *Origanum vulgare* L., essential oil, antimicrobial activity, disc-diffusion method

### ANTIMICROBIAL PROPERTIES OF *ORIGANUM VULGARE* L. FLOWER AND LEAF EXTRACTS

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Antifungal, antimicrobial, insecticidal and antioxidant activities of *Origanum vulgare* L. provide the basis for suggesting that oregano plant extracts may be useful for specific medical conditions. The objective of this study was to determine antibacterial and antifungal activities of methanolic and water extracts from the leaves and flowers of *Origanum vulgare*. Antimicrobial susceptibility testing was done using well diffusion method. Activity of extracts were tested against Gram positive bacteria: *Staphylococcus aureus* ATCC 25923, methicillin-resistant *Staphylococcus aureus* (MRSA) ATCC 33591, *Bacillus subtilis* ATCC 6633, *Enterococcus faecalis* ATCC 29212 and five Gram-negative bacteria: *Salmonella abony* ATCC 6017, *Salmonella enterica* serovar Enteritidis ATCC 31194, *Pseudomonas aeruginosa* ATCC 9027, *Escherichia coli* ATCC 25922, extended-spectrum  $\beta$ -lactamase (ESBL) producing *Escherichia coli* ATCC 35218 and fungi *Candida albicans* ATCC 1023. Antibiotics ampicillin, streptomycin, and antimycotic nystatin were used as positive control. The largest diameters of inhibition zone for methanolic and water flower and leaf extracts were recorded for Gram positive *Bacillus subtilis*, *Staphylococcus aureus*, and methicillin-resistant *Staphylococcus aureus*. Methanolic extracts exhibited antibacterial activity against tested Gram negative bacteria in variable degree while the growth of these bacteria was not inhibited by water extracts. Tested fungi *Candida albicans* was not susceptible to investigated oregano extracts. Therefore it can be concluded that flower and leaf oregano extracts have great antibacterial potential.

**KEYWORDS:** *Origanum vulgare* L., flower and leaf extracts, antimicrobial activity, well diffusion method

### NEW INSIGHTS INTO THE ANTIFUNGAL ACTIVITY OF *TAXUS BACCATA* L. (TAXACEAE)

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*Taxus baccata* L. (Taxaceae), yew tree, is dioecious, small to medium-sized evergreen tree, native to Europe, Africa and Asia, but frequently cultivated worldwide because of its ornamental features. It is long known that all parts of *T. baccata* (except of aril) are toxic, due to the alkaloid taxine. Nevertheless, some parts of yew tree can be used to treat cancer, as well as antimicrobial agents. Unlike the antibacterial properties, potential antifungal activity of *T. baccata* is poorly investigated. In this research, antifungal activity of *T. baccata* methanol and aqueous extracts was tested against three reference strains of fungi: *Candida albicans* ATCC 10231, *Aspergillus brasiliensis* ATCC 16404, and *Ascosphaera apis* MUCL 30769. To assess antifungal effects, agar well diffusion method was used. For extracts preparation, leaves, bark and reproductive structure (berries and microstrobili) are separated for female and male plant respectively. Extraction was performed by 80% methanol and distilled water. Antimycotic solution (penicillin, streptomycin and amphotericin) was used as positive control, while methanol and distilled water were used as negative control. All tests are performed in triplicate. Obtained results showed variable degree of fungal growth inhibition caused by the methanol extracts. Widest inhibition zones (23.33±2.08 and 22.67±0.58 mm) are observed in case of aril extract activity against *A. brasiliensis* and *C. albicans*, followed by inhibition zones in *A. apis* (20.83±0.29 mm) caused by male bark extract. Considering the fact of emerging multidrug resistance of *C. albicans*, interesting finding is inhibition of this strain by all obtained methanol extracts (14.67±1.15 - 22.67±0.58 mm), that is significantly higher in comparison to the activity of standard antimycotic solution (4.90±0.36 mm). Furthermore, according to the available data, male reproductive structures of this species were not tested earlier for their antifungal activity, and this investigation showed relatively high level of fungi inhibition by methanol microstrobili extract. Antifungal effects of yew aqueous extracts were not recorded in this study. Realized research indicates great antifungal potential of *T. baccata*, which should be confirmed by further more comprehensive studies.

**KEYWORDS:** yew, antifungal properties, agar well diffusion method, methanol extracts, aqueous extracts

### ARTEMISIA DRACUNCULUS L. OIL AS POTENTIAL ANTIFUNGAL AGENT

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*Artemisia dracunculus* L. (Asteraceae) is perennial herbaceous plant, native to Europe, South Russia and West Asia. This plant, known as estragon or tarragon, is cultivated in Europe (France, Holland, Hungary) and USA for essential oil production, which is widely used in cosmetic and food industry. The present study describes the chemical composition and antifungal activity of commercial sample of *A. dracunculus* essential oil. Chemical profile of oil was determined by GC-FID and GC-MS techniques. Using macro- and microdilution methods, minimum inhibitory and minimum fungicidal concentrations (MICs/MFCs) were recorded against 19 different micromycetes. In both assays, commercial mycotic bifonazole was used as a control. The oil was rich in monoterpenoids (64.2%), where the main compounds were limonene (34.97%) and *p*-cymene (12.14%). Also,  $\alpha$ -thujene (6.12%), methyl eugenole (4.88%), *trans*-sabinene hydrate (4.18%) and elemicin (3.03%) were present in reasonable amounts. In both applied antifungal tests, oil showed stronger fungicidal potency than bifonazole. Comparison of the results obtained in antifungal tests showed that the most sensitive micromycetes were *Cladosporium cladosporioides*, *C. fulvium*, *Trichophyton mentagrophytes* and *Candida albicans* (MFCs= 0.5-2.5 mg/mL for the oil and MFCs=5-10 mg/mL for bifonazole). The most resistant fungi were *Aspergillus* and *Penicillium* spp. and *Trichoderma viride* (MFCs=10-15 mg/mL for the oil and MFCs=15-20 mg/mL for bifonazole). It is assumed that estragol, which is the most commonly dominant constituent in the tarragon oil, was probably not responsible for the antifungal potency of this sample, since it was present in amount around 1%. Compounds presented in different amounts in the oil acting together and the activity of the oil is due to its possible synergistic effect. Some disadvantages noted in macrodilution method such as huge amount of the oil, low oil dispersion in agar medium, should be overcome by using microdilution method. Presented data suggested that analysed estragon essential oil can be considered as potent antifungal agent which could inhibit the growth of various plant and animal pathogens, food spoilage, mycotoxin producers and other micromycetes.

**KEYWORDS:** *Artemisia dracunculus*, Asteraceae, essential oil, antifungal activity, macrodilution, microdilution

### ANTIFUNGAL ACTIVITY OF PLANT ESSENTIAL OILS AGAINST FUNGI CAUSING GREEN MOULD DISEASE OF CULTIVATED MUSHROOM

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Button mushroom (*Agaricus bisporus* (Lange) Imbach) is the most commonly cultivated basidiomycete worldwide and in Serbia. Biofungicides based on plant oils have advantages compared to chemical fungicides especially considering their harmful effect to the environment. Twenty-two essential oils from Germany and Albania were assayed for inhibitory and fungicidal activity against *Trichoderma* pathogens of cultivated mushroom. The most significant crop losses in mushroom cultivation in Serbia due to green mould are induced by *T. aggressivum* f. *europaeum* Samuels & W. Gams, *T. harzianum* Rifai and *T. atroviride* P. Karsten. Antifungal activity was tested using macrodilution fumigant method. Concentrations of the oils obtained in the air phase were: 0.02, 0.04, 0.08, 0.16 and 0.32  $\mu\text{L mL}^{-1}$  of air with determined minimum inhibitory and fungicidal concentrations. The strongest and broadest activity against all three pathogens was shown by two samples of mint oil (*Mentha piperita* L.) at 0.02  $\mu\text{g mL}^{-1}$ , both from Germany and Albania. *T. atroviride* was the least sensitive to plant essential oils, being susceptible to mint, black pine (*Pinus nigra* L.), cade (*Juniperus oxycedrus* L.), and lavender (*Lavandula angustifolia* Mill.) oils, and all of them were lethal to the pathogen. Ten oils: mint, cade, eucalyptus (*Eucalyptus globulus* Labillardie), lavender, black pine, etc., inhibited the growth of *T. harzianum* and four oils showed fungicidal effects to the pathogen. The most sensitive pathogen was *T. aggressivum* f. *europaeum* as 11 oils (mint, cade, eucalyptus, black pine, lavender, etc.) inhibited growth of the fungi and four oils were lethal. After further *in vivo* experiments, mint essential oil could be incorporated in integrated green mould management.

**ACKNOWLEDGEMENTS:** This study was funded by the Ministry of Education, Science and Technological Development of the Republic of Serbia, Project TR31043 and III 46008.

**KEYWORDS:** essential oils, antifungal activity, cultivated mushroom

### EVALUATION OF GENOTOXICITY AND CYTOTOXICITY OF *HELICHRYSUM ITALICUM* (ROTH) G. DON AND *LAVANDULA ANGUSTIFOLIA* MILL. ESSENTIAL OILS

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Immortelle, *Helichrysum italicum* (Asteraceae) (Roth) G. Don, and lavender, *Lavandula angustifolia* Mill. (Lamiaceae), are aromatic plants used in traditional medicine for centuries due to their medicinal properties. Along with other compounds, special attention is devoted to essential oils that are commercially easily available and used for wide spectrum of diseases and conditions. The main goal of this study was to investigate *in vitro* cyto/genotoxic effects of immortelle and lavender essential oils using plant (*Allium cepa* assay) and animal (human peripheral blood lymphocyte culture) models. Our results showed significantly increased root growth for onion bulbs treated with lavender oil (0.20  $\mu\text{L/mL}$ ) compared to the control group, as well as for bulbs after treatment with immortelle oil (0.20; 0.30  $\mu\text{L/mL}$ ) in comparison with solvent control. In terms of genotoxic effects of lavender on *A. cepa* root meristem cells, our results showed that the frequency of structural chromosome aberrations (sticky chromosomes, chromosomal bridges and vagrant chromosomes) significantly differs from the control. For immortelle, the frequency of sticky chromosomes, chromosomal bridges, chromosome missegregations and multipolar anaphases was significantly higher as compared to controls. By analysing the cytotoxic effects on the plant system, both oils demonstrated increased frequency of apoptotic cells for all concentrations, as well as frequency of necrotic cells (0.10/0.30  $\mu\text{L/mL}$  for lavender/immortelle, respectively). In human lymphocyte culture, statistically significant differences for minute fragments between immortelle oil (0.10  $\mu\text{L/mL}$ ) and controls were observed. Furthermore, significantly increased number of apoptotic cells was detected for the immortelle (0.20  $\mu\text{L/mL}$ ), while both essential oils (lavender at concentrations 0.20; 0.30  $\mu\text{L/mL}$ , and immortelle at all tested concentrations) induced significantly higher number of necrotic cells when compared to the controls. The results of the present study demonstrated that lavender and immortelle essential oils exhibited genotoxic and cytotoxic effects in both plant and animal model systems. Further studies in terms of evaluation of cyto/genotoxic effects of these two essential oils by conducting other mutagenicity tests are needed to strengthen our findings.

**KEYWORDS:** genotoxic and cytotoxic effects, essential oil, immortelle, lavender, *Allium cepa* assay, human lymphocyte culture

### IN VITRO NEUROPROTECTIVE ACTIVITY OF *HYPERICUM PERFORATUM* L. WILD-GROWING PLANTS FROM THE REPUBLIC OF MACEDONIA

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*Hypericum perforatum* L. (St. John's wort) is the most intensively studied medicinal plant due to the presence of a broad range of bioactive compounds with various pharmacological attributes. The aim of this study was to evaluate the contents of total phenolics (TP), flavonoids (TF) and flavan-3-ols (TFL), as well *in vitro* neuroprotective activity of methanolic extracts from roots, non-flower shoots and flower shoots of *H. perforatum* originating from the Republic of Macedonia. The TP, TF and TFL contents in plant extracts were determined according to the Folin-Ciocalteu, aluminium chloride and 4-dimethylaminocinnamaldehyde colorimetric methods, respectively. The *in vitro* neuroprotective activity of methanolic extracts was evaluated through testing their capacity for inhibition of acetylcholinesterase (AChE) linked to Alzheimer's disease and tyrosinase (TYR) related to Parkinson's disease. Present results demonstrated that TP contents in non-flower shoots and flower shoots were about 2.9-fold higher than that of roots. In comparison to root extracts, flower shoots and non-flower shoots showed 2.4 and 2.7-fold higher TF concentrations, respectively. Similar amounts of TFL were observed between both aerial plant parts, which were approximately 2.5-fold higher compared to roots. The *H. perforatum* extracts displayed considerable and dose-dependent inhibition of AChE activity. Flower shoot extracts exhibited the highest AChE inhibition (59.6%), followed by roots (53.9%), while non-flower shoots had the lowest enzyme inhibition (46.1%) at 250  $\mu\text{g}\cdot\text{mL}^{-1}$ . Also, flower shoots showed about 1.3-fold higher AChE inhibition at 150  $\mu\text{g}\cdot\text{mL}^{-1}$  compared to root and non-flower shoots. The AChE inhibitory activity upon dose of 50  $\mu\text{g}\cdot\text{mL}^{-1}$  did not differ significantly among tested samples. The TYR inhibition by flower shoots (from 83% to 87%) and non-flower shoots (from 80 to 90%) was significantly higher compared to roots (from 55% to 69%). This study indicated that *H. perforatum* extracts possess significant AChE and TYR inhibitory activities, which might be contributed to the accumulation of phenolic compounds with neuroprotective properties. Therefore, *H. perforatum* could be potentially used for the treatments of patients with neurodegeneration and cognitive abnormality.

**KEYWORDS:** *Hypericum perforatum* L., neuroprotective activity, phenolic compounds

### VIABILITY REDUCTION OF TUMOR NFS-60 CELLS BY LUTEOLIN

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Plant flavonoids are natural, polyphenolic compounds frequently present in human diet. Luteolin (3',4',5,7-tetrahydroxyflavone) is a yellow pigment from flavones class, present in the plants with a yellow flowers, other fruits and vegetables such as *Daucus carota* L., *Ficus carica* L., but also in the leaves of green condiments e.g. *Origanum vulgare* L., *Petroselinum crispum* (Mill.) Fuss. Antitumor properties of luteolin are widely known as well as a various protective effects including antioxidant and antimutagenic activity, inhibition of genotoxic damage etc. As luteolin is confirmed inhibitor of some cancer cell proliferation, its antiproliferative effects were tested in NFS-60 tumor cell line. NFS-60 is a tumor cell line of murine myeloblastic leukemia, with high proliferation and differentiation capacity. NFS-60 cells were cultivated at 37° C in RPMI 1640 medium supplemented with L-glutamine, FBS and antibiotics. After 24 hours, cultures were treated with three concentrations of luteolin (5, 10 and 20  $\mu\text{M}$ ) and incubated for further 48 hours (Nuve Incubator, EC 160). Positive, negative and blank controls were set as well. The parallel experiment was conducted on peripheral blood lymphocytes, with the additional concentration of 50  $\mu\text{M}$  tested as well. The TransDetect® Anenexin V-EGFP/PI assay was performed for detection of apoptotic and necrotic cells induced by luteolin. Percentage of cell viability was calculated using trypan blue exclusion assay. Following luteolin treatment, early and late apoptotic and necrotic tumor cells were clearly distinguished on Olympus BX51 epi-fluorescent microscope. Cell viability declined from the 90.8 % to 59.4 % with the increase of concentrations. Increase of non-viable tumor cells was dose-dependent. The high luteolin concentration of 50  $\mu\text{M}$  applied in lymphocytes cultures, comparable to lower in tumor cells, revealed high viability percentage (73,9 %). Obtained results indicate the potential of luteolin to reduce viability of NFS-60 tumor cells contributing to intense research of anti-cancer activity.

**KEYWORDS:** tumor cells, luteolin, trypan blue, growth inhibition

### BIOACTIVE PROPERTIES OF *HERACLEUM SPHONDYLIIUM* L. SUBSP. *TERNATUM* (VELEN.) BRUMMITT. (APIACEAE) EXTRACTS

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In last decades, the uses of plant or plant products have significantly increased because chemical products are very expensive and have many unfavorable health effects. In addition, several studies have suggested that there is negative correlation between the consumption of the plant products and the incidence of some chronic diseases such as atherosclerosis, Alzheimer's diseases and diabetes mellitus. This phenomena lead to the emergence of the terms of "nutraceuticals" to indicate the functional food able to prevent or facilitate the treatment of such diseases. *Heracleum* (Apiaceae) species, also known as hogweed, are traditionally used as food additives, spices, and flavoring agents. From this point, antioxidant capacity and enzyme inhibitory effects of three extracts (ethyl acetate, methanol and water) from *Heracleum sphondylium* subsp. *ternatum* aerial parts were tested. Antioxidant capacity was assessed with different assays including free radical scavenging (ABTS and DPPH), reducing power (FRAP and CUPRAC), phosphomolybdenum and metal chelating assays. Enzyme inhibitory effects were tested against cholinesterase, tyrosinase, amylase and glucosidase. The water extract exhibited stronger antioxidant capacity compared to ethyl acetate and methanol extracts. Also, the water extract had remarkable anti-tyrosinase activity. However, the ethyl acetate and methanol extracts were more active on cholinesterase than water extract. These findings suggest that *Heracleum sphondylium* subsp. *ternatum* may be assisted as a source of biologically-active compounds for designing novel functional food and drug formulations.

**KEYWORDS:** *Heracleum sphondylium* subsp. *ternatum*, enzyme inhibitor, antioxidant, Alzheimer, Diabetes mellitus

### INVASIVE ALIEN PLANTS AS A SOURCE OF BIOLOGICALLY ACTIVE COMPOUNDS

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According to the novel weapon hypothesis, allelopathy is one of the possible mechanisms that enable alien plants to become invasive in the newly colonized areas. The allelopathic effects are seen as the inhibition of seed germination and/or growth and development of the nearby plants. The allelochemicals are actively released from roots or degrading plant material and affect other plants directly or indirectly (via soil microbiota). Invasive alien plants (IAPs) therefore successfully colonize and spread in new habitats. By removal of IAPs we can collect high quantities of their biomass which presents a potential source for the extraction of biologically active material. To evaluate the potential of IAPs for preparation of biologically active compounds, invasive knotweed (*Fallopia japonica*) and goldenrods (*Solidago canadensis* and *S. gigantea*) were collected and used for preparation of extracts and essential oils which were tested for antifungal and antibacterial activity using standard growth inhibition tests. The results show that preparations of goldenrods were more effective than from knotweed and that essential oils had stronger inhibitory effects than extracts, and root extracts were more effective than shoots. Gram positive bacterial strains were more susceptible to IAPs than Gram negative. For the antifungal activity, the growth of all selected species was affected when fungi were exposed to IAPs. Additionally, the IAPs were tested as potential fungicides for the treatment of wheat grain to prevent crop spoilage. To conclude, selected IAPs show a potential to use their biomass for preparations that control bacterial and fungal infections.

**KEYWORDS:** *Fallopia*, *Solidago*, extract, antibacterial activity, antifungal activity, growth inhibition

### DELIGNIFICATION OF THE MOST COMMON AGRICULTURAL PLANT RESIDUES BY *PLEUROTUS PULMONARIUS*

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Agricultural plant residues are a widespread lignocellulosic biomass source which commonly retains as a ballast in nature although it can potentially be used for bioethanol produc-

### BALKAN COASTAL HALOPHYTES: POTENTIAL SOURCE OF BIOLOGICALLY ACTIVE COMPOUNDS

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In order to overcome unfavourable environmental conditions, halophytes evolved ability to synthesize a range of different secondary metabolites, mainly phenolics. Some halophytes are used in the folk medicine, while for the others the ethnobotanical data are scarce. A comparative analysis of phenolic compounds and related antioxidant and antimicrobial activity was performed on group of under-researched coastal halophytes from the region of the Southeast Europe. Total phenolics, flavonoids, tannins and anthocyanins were determined in methanolic and hot water extracts (tea infusions) from 24 halophytic species sampled from different coastal saline habitats of three Mediterranean countries – Montenegro, Albania and Greece. The list of studied halophytes includes: the *Panocratium maritimum*, *Salsola kali*, *S. soda*, *Salicornia europaea*, *Sarcocornia fruticosa*, *Halimione portulacoides*, *Halocnemum strobilaceum*, *Limonium angustifolium*, *L. vulgare*, *Polygonum maritimum*, *Tribulus terrestris*, *Medicago marina*, *Euphorbia peplus*, *Cakile maritima*, *Alkanna tinctoria*, *Centaurium maritimum*, *Vitex agnus-castus*, *Calystegia soldanella*, *Echinophora spinosa*, *Eryngium maritimum*, *Artemisia maritima* and *Xanthium italicum*. The content of the total phenolic compounds ranged from 13.23 to 376.08 mg of GA/g of extract, and 33.68 to 511.10 mg/ml for infusion samples, respectively. Content of flavonoids ranged from 12.63 to 77.36 mg of Ru/g in the methanol extracts, and from 12.13 to 26.35 mg/ml in infusion. Total tannins varied from 0.05 to 2.44 g/l, while the concentration of anthocyanins was in range of 1.31 - 39.81 mg/l. The obtained values for antioxidant activity of methanolic extracts ranged from 1147.68 to 15.02 µg/ml and from 1613.05 to 21.96 µg/ml for tea infusions. The best antioxidant properties were obtained for *Polygonum maritimum* and *Limonium vulgare*. Antimicrobial activities were determined using microdilution method and were tested against *Staphylococcus aureus*, *Bacillus cereus*, *Sarcina lutea*, *Klebsiella oxytoca*, *Pseudomonas aeruginosa*, *Salmonella enteritidis* and *Aspergillus brasiliensis*. Investigated extracts exhibited various activities with MIC ranging from 0.015 to 20.00 mg/ml. Significant antimicrobial effects was found for extracts from *Limonium vulgare*, *L. angustifolium* and *Artemisia maritima*. According to obtained results, some of studied coastal halophytes can be considered as rich natural sources of phenolic compounds,

especially flavonoids with favorable antioxidant and antimicrobial properties.

**KEYWORDS:** coastal halophytes, Southeast Europe, phenolics, secondary metabolites, antioxidant and antimicrobial activity

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### RUBUS ULMIFOLIUS – FREE RADICAL SCAVENGING AND METAL CHELATING ACTIVITY

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*Rubus ulmifolius* Schott (Rosaceae) is a well known shrub extending from Europe to Siberia. It is a wild-growing plant, widespread on hills and mountains and also cultivated as acrop. Dewberries are consumed worldwide because of their taste and health benefits. *Rubus* spp. have been used in traditional medicine and their extracts possess anti-inflammatory, antiviral and antimicrobial effects, antiproliferative activity against cancer cells and wound-healing properties. Free radical scavenging activity of the ethanol extracts from leaf, stem and fruit of *R. ulmifolius* were estimated against 2, 2-diphenyl-1-picrylhydrazil (DPPH) and 2,2-azino-bis (3-ethylbenzothiazoline-6-sulfonic acid) (ABTS) assays. Metal chelating activity was measured by ferric reducing ability (FRAP). Total contents of phenols and flavonoids were also determined. Total phenol content in the extracts was determined using Folin-Ciocalteu reagent and their amounts ranged between 35.99 for fruit extract to 353.07 mg gallic acid per g (GAE/g) for stem extracts. The amounts of flavonoids varied from 3.15 mg quercetin equivalent per g (QE/g) for fruit extract to 34.38 mg QE/g for leaf extract of *R. ulmifolius*. The highest antioxidant activity against DPPH and ABTS radicals expressed as IC<sub>50</sub> showed stem extract (0.085 mg/mL and 0.172 mg/mL). Given values for FRAP varied between 0.228 for fruit extract to 2.708 μmol Fe/mg for the stem extract. In all applied methods stem extracts showed the best activity. Further research is needed to identify individual components from *R. ulmifolius*, which form antioxidative system, because of their applications in functional food and/or pharmaceutical industries.

**KEYWORDS:** *R. ulmifolius*, phenols, flavonoids, DPPH, ABTS, FRAP

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### GERMINATION INHIBITION BIOASSAY OF EXTRACTS AND ESSENTIAL OILS FROM PLANT SPECIES

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Synthetic herbicides have harmful effects on the environment and human health. Therefore, the establishment of natural products with herbicidal activity is very important. Methanolic extracts (aerial parts of *Galanthus elwesii* Hook.f., *Geranium sanguineum* L. and *Tanacetum vulgare* L.; leaves of *Leucocjum aestivum* L., *Salvia officinalis* L. and *Narcissus pseudonarcissus* cv. Carlton), acetone exudates (aerial parts of *Artemisia absinthium* L., *Salvia sclarea* L. and *Tanacetum vulgare*, leaves of *Salvia officinalis* L.) and essential oils (aerial parts of *Pelargonium graveolens* L'Hér., *Origanum heracleoticum* L., *Artemisia absinthium* L. and *Thymus longedentatus* (Degen & Urum.) Ronniger; leaves of *Salvia officinalis*,) were assayed as germination inhibitors. *Lolium perenne* L., *Trifolium repens* L. and *Trifolium pratense* L. were used to develop the germination inhibition bioassay. Methanolic extracts and acetone exudates of studied species were applied as aqueous solutions at 1 mg/mL concentration. Essential oils were tested as aqueous solutions at 5 μL/mL. Hundred seeds of the three weeds were placed on filter papers moistened with the solutions cited above and incubated at room temperature for 7 days. At the end of this period the rate of germinated seeds was estimated. Among the tested samples the essential oils of *Origanum heracleoticum* showed the most significant germination inhibitory activity. The essential oil had inhibitory effect on the three tested weeds. No germination of seeds of *Lolium perenne* and *Trifolium repens* was detected, and the germination rate in *Trifolium pratense* was very low (6.5 %). Methanolic extract of *Narcissus „Carlton”* and essential oils of *Thymus longedentatus* showed specific activity against *Trifolium repens* (3% of germination estimated) and *Lolium perenne* respectively (0% of germination estimated). Chemical composition of active essential oils and extract of *Narcissus „Carlton”* were analyzed by GC/MS. The main component of *Origanum heracleoticum* was determined as carvacrol. Galantamine, hemantamine and tazetine were identified as main alkaloids of *Narcissus „Carlton”*. The essential oil of *Thymus longedentatus* is rich of citral isomers. The received results showed that essential oils of *Origanum heracleoticum* and *Thymus longedentatus* as well as methanolic extract of leaves of *Narcissus „Carlton”* are promising sources of bioherbicidal activity.

**ACKNOWLEDGEMENTS:** The authors are grateful for the financial supported by the Bulgarian National Science Fund, Bul-

garian Ministry of Education and Science (Grant DN 16/2/ from 11.12.2017).

**KEYWORDS:** Amaryllidaceae, Asteraceae, Lamiaceae, Geraniaceae, germination inhibition

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### FINDING OUT THE RIGHT RUBUS - ISOLATION OF DNA FROM COMMERCIALY AVAILABLE PLANT MATERIAL

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The genus *Rubus* (Rosaceae) consists of more than 750 species, but only a handful of those have been domesticated. Leaves and fruits of *Rubus* are used in traditional medicine for their antimicrobial, anticonvulsant, muscle-relaxant and radical scavenging activity, but recent research also showed anti-inflammatory and antiviral effects, antiproliferative activity against cancer cells, antitumor and wound-healing properties. Since the composition and the abundance of these compounds is genetically determined, identification of populations that have been proved to have significant biological activity is of imperative. However, plant material commercially available can rarely be identified correctly using traditional taxonomic tools due to extremely high morphological variability which is characteristic for species of this genus, so molecular markers could prove to be useful. During the last few decades, molecular markers have been used to estimate the genetic diversity of *Rubus* populations. Studies focusing on the genetic diversity of *Rubus* spp. have been conducted using various types of molecular markers, including SSRs. The objective of this preliminary research was to optimise isolation procedure of DNA from fruits and leaves not prepared for molecular analyses but for commercial sale, and to identify potential SSRs useful for germplasm identification using *in silico* analysis. Seeds from frozen fruits and leaves dried in dark at room temperature of 6 wild-growing species and 6 commercially available cultivars were used as a source of DNA. Five different CTAB protocols were used, focusing on removal of high concentrations of polyphenols from leaves, and proteins from seeds. The quality of DNA was assessed using spectrophotometric analysis, gel electrophoresis and PCR amplification of two different plastid regions (small 350bp *trnL-trnE*, and 1.5kbp *rps16-trnK*). The best DNA quality was obtained in protocols D and E, using high concentrations of NaCl, 1.4 M CH<sub>3</sub>COONa and 0.5% β-mercaptoethanol. Even though there has been some degradation of DNA in leaves, PCR products were obtained both in leaves and seeds. *In silico* analysis of SSRs showed the potentially large number of SSRs that can be used in the studied species. According to our analysis, cross-species utilization of

already developed SSRs is potentially possible.

**KEYWORDS:** *Rubus*, cultivars, wild populations, seeds, leaves, DNA isolation, SSRs

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### THE ECONOMIC IMPORTANCE OF PLANT SPECIES OF SALT STEPPES AND SALT MARSHES IN BAČKA AND BANAT REGION

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Pannonic salt steppes and salt marshes belong to the most threatened European habitats. This specific eco-systems are characterized by zonation of vegetation, with dominant salt-tolerant grasses and herbs that tolerate or even demand salt concentrations in the soil water. As such, these types of grasslands were not the most favorable targets of arable farming, but large areas affected by mineral fertilization, drainage, soil melioration and/or commercial seeding. However, many of economic valuable plants can be found in Pannonic salt steppes and salt marshes. The aim of this paper is to point out economic value of salt steppes and salt marshes of Bačka and Banat region by analyzing species found on this habitat. In this paper we analyzed 377 taxa from saline habitats which belong to two different classes of vegetation – *Thero-Salicornietea* Tx. in Tx. et Oberd. 1958 and *Festuco-Puccinellietea* Soó ex Vicherek 1973. All taxa are exported from relevés deposited in database using Turboveg for Windows ver 2.92a software. Economic value of taxa were assumed according GRIN-Global database criteria (<https://npgsweb.ars-grin.gov/gringlobal/taxon/taxonomysimple.aspx>). Results of analysis are distributed in 37 categories. Results of analysis of saline flora show that the largest number of taxa (87) can be used in medicine or folklore. Second largest category (65 taxa) can be harmful for mammals. Large number of taxa belong to categories which are important as food source for animals. Categories with negative economic importance are also present. The largest category of taxa with negative economic importance are potential seed contaminants, with 123 taxa. Also, there are 43 taxa assumed as weeds. The last two groups could be threatening factors for Pannonic salt steppes and salt marshes in Bačka and Banat region in the future. Taxa with positive and negative economic importance can have endangering effect on salt steppes and salt marshes. Strong presence of both groups in results of this paper indicates the need to enhance the protection of these fragile habitats, their further evaluation and ongoing monitoring.

**KEYWORDS:** saline vegetation, economic importance, negative economic importance

Poster presentation 66 06 76

### ASSESSMENT OF THE STATUS OF MEADOWS OF VOJVODINA THROUGH THE ASPECT OF SUSTAINABLE EXPLOITATION

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Anthropogenic effect on the vegetation is important factor which influences the course of successive changes, but also brings the risk of endangered habitats collapse, by changing the state of those plant communities that bring the greatest benefit to the local communities and human population. Some secondary phytocenoses are directly dependent on human influence and without certain management, they would disappear forever in natural succession. The group of secondary phytocenoses includes meadows and pastures. Meadows and pastures of the class Molinio-Arrhenatheretea Tx. 1937 can be found on fertile and deep substrates, from the lower to medium altitudes, in the river valleys, forests glades, abandoned clear cuts, on the loess profiles and between sand dunes. Within the Vojvodina Province, this class includes four orders, seven alliances and about 50 associations. The aim of this paper is to assess the status of Vojvodina meadows through the aspect of their sustainable exploitation, by integrating biodiversity and ecosystem services in order to manage and use natural resources. More than 800 phytocenological records of the class Molinio-Arrhenatherete Tx. 1937 were analyzed. Each phytocenological record was analyzed in accordance with the participation of taxa that are of economic importance. An estimation model of phytocenosis assessment was established in accordance with the fidelity and average coverage of economically important species. The assessment model was modified in accordance with the conservation status of phytocenosis and the presence of rare and protected taxa. The largest number of taxa occurring in meadow phytocenoses is characterized by medicinal properties, slightly less represented by honey plants, then decorative plants, but also species of negative economic importance, especially a group of weeds and invasive species. Since meadows are found in the midst of intensive agriculture, the consequences of degradation are almost in every case present. Uncontrolled grazing is a threat to the diversity of wet meadows. Moreover, xerophytic vegetation is intensely mowing in order to meet the needs of modern livestock breeding. The economic wealth of this type of vegetation poses a threat to its survival and plans need to be implemented on the sustainable use of resources and sustainable use of these phytocenoses.

**KEYWORDS:** TurboVeg ver 2.292a, Molinio-Arrhenatheretea, ecosystem services

Poster presentation 67 06 69

### A PRELIMINARY INVESTIGATION ON THE ELEMENTAL COMPOSITION OF POLLEN SAMPLES FROM HAKKARI, TURKEY

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Pollen contains very valuable nutrients for human metabolism. It is rich in vitamins and minerals, with a high level of protein and carbohydrate sources. The chemical composition of the pollen differs depending on the geographical and floral origins of the region from which it is obtained. Many plants are used as a source of pollen by bees. For this reason, the mineral composition of pollen samples from different region also vary. In this study, it was aimed to determine the elemental composition of pollen samples collected from eight different regions of Hakkari, Turkey. For this purpose, the concentrations of the elements Li (Lithium), Be (Beryllium), B (Boron), Na (Sodium), Mg (Magnesium), Al (Aluminum), Si (Silicon), P (Phosphorus), K (Potassium), Ca (Calcium), V (Vanadium), Cr (Chromium), Mn (Manganese), Fe (Iron), Co (Cobalt), Ni (Nickel), Cu (Copper), Zn (Zinc), Ga (Gallium), As (Arsenic), Se (Selenium), Rb (Rubidium), Sr (Strontium), Ru (Ruthenium), Rh (Rhodium), Pd (Palladium), Ag (Silver), Cd (Cadmium), In (Indium), Sn (Tin), Sb (Antimony), Te (Tellurium), Cs (Cesium), Ba (Barium), Hf (Hafnium), Ir (Iridium), Pt (Platin), Au (Gold), Hg (Mercury), Tl (Thallium), Pb (Lead) and Bi (bismuth) of the pollen samples were measured at the ppb level using ICP-MS. As a result, the concentrations of K, P, Mg, Si, Ca, Al and Fe elements were found to be the highest in the studied samples. These results provide the first detailed data on the elemental composition of pollen samples collected from the Hakkari province.

**KEYWORDS:** pollen, elemental composition, ICP-MS

Poster presentation 68 06 43

### BOTANICAL CHARACTERIZATION OF HONEY FROM BOSNIA AND HERZEGOVINA

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The present study reports first detailed botanical characterization of honey from the Bosnia and Herzegovina. Hundred samples from different ecological and botanical regions were analyzed during the three vegetation seasons (2015-2017).

The qualitatively-quantitative analysis of melisoplanological slides was in accordance with the methodology described in rulebook on methods for controlling honey and other bee products of Bosnia and Herzegovina. Melisoplanological profiles were prepared for each sample/slide. All the elements determined in the palinomorphological profiles represent a unique "imprint" of the environment from where the honey originates. The dominant and accompanying honey plants species were determine, and the samples were qualified as monofloric and polyphloric. All monofloral honey samples considering the dominant species are divided into botanical categories, of which the most commonly determined were: *Robinia pseudoacacia*, *Tilia* sp., *Castanea sativa* and *Salvia officinalis*. Polifloric honey is divided into meadows and forest honey. Based on the micromorphological ornamentation, 30 000 pollen grains were determined. A total of 25 families were found, and the most common species were from families: Fabaceae, Fagaceae, Asteraceae, Rosaceae, Oleaceae, etc.

**KEYWORDS:** pollen, honey, melissopalynology, botany

Poster presentation 69 06 68

### HONEY PLANTS OF TURKEY, BAYBURT

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Turkey is one of the world's most important countries in terms of beekeeping and beekeeping is carried out in every province of Turkey as a traditional agricultural activity. One of these provinces, Bayburt is located in Turkey's Eastern Black Sea region and has a very important position in terms of beekeeping. In this study, the plants important for beekeeping in Bayburt province and the yields of nectar and pollen of these plants were evaluated. As a result, taxa belonging to families such as Apiaceae, Asteraceae, Boraginaceae, Brassicaceae, Fabaceae and Lamiaceae were observed in Bayburt province. Especially plants belonging to the taxa of *Alkanna* spp., *Anchusa* spp., *Aster* spp., *Astragalus* spp., *Carum* spp., *Centaurea* spp., *Cerinth* spp., *Crepis* spp., *Echinops* spp., *Eryngium* spp., *Isatis* spp., *Lamium* spp., *Medicago* spp., *Melilotus* spp., *Myosotis* spp., *Onobrychis* spp., *Onosma* spp., *Pimpinella* spp., *Taraxacum* spp., *Thymus* spp., *Trifolium* spp. have been determined to be important for beekeeping in this province. It has been observed in the literature that these taxa have pollen or nectar yields at dominant, secondary, minor or trace levels. This indicates that the potential for beekeeping is high because of the rich floristic structure of Bayburt.

**KEYWORDS:** honey plants, floral origin, nectar and pollens

Poster presentation 70 06 09

### HONEY: EFFECT OF EXTRACTION TYPE ON PHENOLIC CONTENT AND ANTIOXIDANT CAPACITY RESULTS

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Methodological approach for determination of phenolic compounds and antioxidant capacity of honey differs between research papers. Extraction process is an important step in the evaluation of total phenolics and antioxidant capacity, but most of the available data on phenolic composition of honey originate from spectrophotometric methods performed without extraction, directly on fresh honey samples, often with different solvents. In this study we screened for the possible differences in total phenolics and antioxidant capacity results depending on the extraction type to which the honey was subjected. Experiments were performed using honey of *Castanea sativa*, *Tillia* spp. and *Robinia pseudoacacia* species, in triplicates for each honey sample - (a) fresh honey dissolved in water, (b) fresh honey dissolved in ethanol and (c) ethanol extract prepared using column chromatography. In most cases, the results acquired with ethanol extract obtained by column chromatography had significantly higher readings (for example *C. sativa*: ABTS (42.1±2.1%), DPPH (48.6±0.7%), FRAP (91.9±0.4, total phenolics (160.1±12.9 mg galic acid equivalents/100 g of honey)) then the results acquired with both dilutions of fresh honey samples (*C. sativa*: dissolved in water - ABTS (24.2±0.4%), DPPH (24.6±1.5%), FRAP (81.3±0.2%), total phenolics (67.1±5.0 mg galic acid equivalents/100 g of honey); *C. sativa* dissolved in ethanol - ABTS (16.2±4.8%), DPPH (3.0±0.2%), FRAP (78.5±0.7%), total phenolics (44.1±9.2 mg galic acid equivalents/100 g of honey). Among the fresh honey samples, no strict tendency could be observed, in some cases water was better solvent, but in another ethanol was better (for example total phenolics in *R. pseudoacacia* were 14.9±4.2% for water dissolved honey, and 49.7±17.6% for ethanol dissolved honey, while FRAP was 44.7±0.1% for water dissolved honey, and 30.2±1.8% for ethanol dissolved honey). Furthermore, total phenolic content and antioxidant capacity results do not completely match between fresh honey samples and those extracted using column chromatography. These results imply that omission of extraction step using column chromatography reduces the sensitivity and specificity of spectrophotometric methods for assessment of total phenolics and antioxidant capacity of honey.

**KEYWORDS:** antioxidants, column chromatography, methodology, polyphenols, spectrophotometry



## Session 7. Agronomy and Forestry



Balkan  
Botanical  
Congress

Introductory lecture 01 07 12

### ADAPTIVE VARIATION IN PHYSIOLOGICAL, ANATOMICAL AND MORPHOLOGICAL TRAITS OF EUROPEAN BEECH POPULATIONS IN SOUTHERN EUROPE

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European beech (*Fagus sylvatica* L.) is one of the most abundant tree species in Europe. Due to high genetic diversity and phenotypic plasticity, beech is well adapted to different climatic conditions within its natural range. Climatic changes are projected to negatively affect beech populations in central and south-east Europe, where growth decline and increased mortality of beech trees are expected to occur as the consequence of increased frequency and severity of heat stress and drought. For this reason, the assessment of adaptive potential of beech populations has a double significance: 1) to provide an insight into the capacity of beech populations in south-east Europe to respond to climatic changes, and 2) to identify suitable provenances that could be used as seed sources for future reforestation programmes in Europe. In this paper we present the results on adaptive variation of different physiological, anatomical and morphological traits in European beech grown in natural populations, as well as populations cultivated in common garden experiments. The results are based on numerous experiments that are conducted at the Institute of Lowland Forestry and Environment and their partner institution during several years, and which involved more than 30 beech populations, covering both core and marginal sites of the species distribution range. The research on variation in xylem vulnerability to embolism in beech populations clearly evidenced that the southernmost beech populations growing in a warmer drier climate and with lower habitat suitability have higher resistance to embolism than those from Northern Europe growing more favourable conditions. Similarly, the results from common garden experiments demonstrated that certain populations originating from drier sites showed higher physiological performance during the heat wave compared to the populations locally adapted to mesic site conditions. Likewise, the results from the same field experiments indicated the presence of notable genetic variability between different populations that could have a substantial effect on species adaptive capacity. Finally, the results evidenced the possibility of plastic populations' response to altered environmental conditions in terms of certain leaf and wood anatomical traits.

**KEYWORDS:** European beech, climate change, adaptation, functional traits

Oral presentation 02 07 10

### THE EFFECT OF TEMPERATURE AND PRECIPITATION ON THE RADIAL GROWTH OF ORIENTAL BEECH IN THE AMANOS MOUNTAINS (SOUTHERN TURKEY)

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Oriental beech (*Fagus orientalis* Lipsky) has a distribution range from the Balkans in the west, through Anatolia (Asia Minor), to the Caucasus, northern Iran and Crimea. In Turkey, the species is mainly distributed in the Black Sea Region, Thrace and the south of Marmara Sea. However, the isolated relict populations of the species also occur on the Amanos Mountains in the Eastern Mediterranean Region of Turkey. These populations are known as the most southerly populations within its geographical distribution range. Oriental beech forms both pure stands and mixed forests with conifers and other deciduous broadleaf species. This study was carried out to construct a site chronology of oriental beech in Topaktas site of the Amanos Mountains, one of its most southerly populations, and to investigate the relationships between tree-ring width and some climate variables such as the monthly maximum, minimum and mean temperature and monthly total precipitation. Ring widths were measured on 33 increment cores taken from 17 oriental beech trees in the study site, and 53-year-long tree-ring chronology were built for the period of 1961 - 2013. Pearson correlation coefficients were used to investigate relationships between radial growth and climate. The radial growth of oriental beech in the site was found to be sensitive to climate with mean sensitivity value of 0.18. The results showed that high precipitation in June leads to produce wider rings ( $r = 0.39$ ,  $p < 0.01$ ), while maximum temperature in the same month has a negative significant effect on radial growth ( $r = -0.32$ ,  $p < 0.05$ ). On the other hand, high precipitation in February decrease ring width of oriental beech ( $r = -0.30$ ,  $p < 0.05$ ) in Topaktas Site of the Amanos Mountains (Southern Turkey).

**KEYWORDS:** *Fagus orientalis* Lipsky, dendroecology, tree-rings, climate

Oral presentation 03 07 07

### THE IMPACT OF *AMBROSIA ARTEMISIIFOLIA* ON THE VEGETATIVE PRODUCTION OF *AMBROSIA TRIFIDA*

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In Serbia, *Ambrosia artemisiifolia* (AA) is present in high densities and it reduces crop yields. In addition to AA, *Ambrosia trifida* (AT) is locally present in Serbia and it is expected that it will be more competitive than AA once it occupies the same area. The aim of this study was to determine the biomass of AT under competitive interaction with AA. The experiment was conducted using a replacement design model in ratios plants AA:AT (%) as: a-100:0; b-80:20; c-40:60; d-60:40; e-20:80 and f-0:100, in a completely randomized block design, with four replicates. The experiment has two parts where the plants coexist at a low density (10 plants/m<sup>2</sup>) and high density (100 plants/m<sup>2</sup>). The vegetative parameters of AT were measured in three assessments during 2016 and these results were statistically analyzed. The height of plants was higher in all treatments in high density, when compared with low density. In two densities (low and high), the plants from the b treatment reached their maximum heights in all assessments. Statistically significant differences were found between low and high density in treatments c, d and e (1<sup>st</sup> assessment) and d and f (2<sup>nd</sup> and 3<sup>rd</sup> assessment). The average width of plants and number of leaves was higher in low density when compared with high density. Statistical differences were found between c, e and f treatments (1<sup>st</sup> assessment), c, d and f (2<sup>nd</sup> assessment) and f (3<sup>rd</sup> assessment) when comparing the width of plants in low and high densities and in all treatments (2<sup>nd</sup> assessment) when estimating the number of leaves. The average dry weight values ranged between 5.8-8.99 g (1<sup>st</sup> assessment), 7.4-13.01g (2<sup>nd</sup> assessment) and 10.01-13.54g (3<sup>rd</sup> assessment) in low density. The values of dry mass in high density ranged between 6.61-8.38g, 8.66-11.79g and 9.88-16.13 g respectively. The results show that the vegetative mass of AT in high densities is higher compared with low densities (as a result of a more pronounced light competition). The obtained results have shown the competitive capacity of AT and its invasiveness in coexistence with AA.

**ACKNOWLEDGEMENTS:** Ministry of Education, Science and Technological Development of the Republic of Serbia (Project III46008).

**KEYWORDS:** competition, replacement design model

Oral presentation 04 07 01

### WHY CULTIVATE PLANTATIONS OF WILLOW?

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The short rotation coppice plantation of trees, like willow, poplar and some others, is one of the most important ways of bioenergy production in agroforestry which is a sustainable way of land management which integrates both agricultural and forestry practices. Easy vegetative propagation by cuttings and the production of a large number of new shoots leads to high biomass production in a short period of time. Therefore, willows are commonly used as an energy crops with high yield of biomass. Genus *Salix* L. (willows) is the largest genus of the family Salicaceae with about 450 species with relatively high adaptability to different ecological conditions and genetic intraspecific variability, as a result of a large number of intraspecific hybridizations and differentiations. According to Flora Serbia, there are 18 indigenous species of *Salix* genus growing in Serbia, which represent a great genetic potential for creation of genotypes suitable for high and fast wood production in specific agro-technical conditions. In order to define maximal potential of organic (wood) production of willow plants, plantation has been established with two-year old plants (shoot/root: 2/3, 2/2 and 2/0) and one-year seedlings (shoot/root: 1/2, 1/1 and 1/0) on soil with optimal fertilization and irrigation, as well as with appropriate plant diseases and weed control. The best average density of seedlings was 15 625 cuttings per hectare. On the basis of our results and many literature data it could be concluded that this type of willow plantation can last and be used for 20 to 25 years with a harvest period every three years and yield of wood biomass achieving 10-15 tons of dried wood per hectare. There are many benefits of short rotation coppice plantation of willow for energy production, in particular: a renewable source of clean energy; high energy value (1kg of dry biomass ~ 1kg of coal = 0.5l of grease = 0.5m<sup>3</sup> of methane); easy to use and storage (1m<sup>3</sup> = 650 kg); insignificant amount of pollutants is released by combustion, with ash content less than 1%.

**KEYWORDS:** plantation, *Salix* sp., yield of biomass, willows

Poster presentation 05 07 11

### RESPONSES OF MORPHOLOGICAL LEAF TRAITS TO LONG-LASTING DROUGHT IN PEDUNCULATE OAK PROVENANCES

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Pedunculate oak (*Quercus robur* L.) is economically one of the most valuable hardwood tree species. Due to its wide ecological niche the species forms different types of forests widespread in Europe. In the context of an increased risk of future extreme drought events across Europe, the capacity of pedunculate oak to acclimate to long-lasting drought, deserves further attention. As leaf morphology is tightly linked to plant water relations it attracted much attention in terms of acclimation and adaptation of forest trees to drought. Since spring 2015, for three successive years pedunculate oak samplings originating from four European (Estonia, Lithuania, Poland and Hungary) and four Croatian provenances (Repaš, Koška, Otok and Karlovac) were exposed to long-lasting droughts (from spring to mid-summer) and afterwards rewatered. During the growing season of 2018 fully developed and healthy leaves from each plant were sampled and analyzed. Leaf morphology of samples from studied provenances responded to long-lasting droughts in different ways. Generally, morphological changes indicate drought acclimation capabilities of oak, but there was strong differentiation among provenances which indicates adaptedness to their original habitat conditions.

**KEYWORDS:** *Quercus robur*, morphological traits, drought stress, acclimation, provenances

Poster presentation 06 07 22

### RESPONSE OF BLACK POPLAR CLONES CUTTINGS TO DROUGHT STRESS

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Drought represents one of the most significant plant stressors and has important effects on plants and ecosystem functioning. Plants have evolved complex physiological and biochemical adaptations to adjust and adapt to a variety of environmental stresses. In the present study we investigated the antioxidative response of leaves of black poplar clones B-229,

PE 19/66 (*Populus deltoides*) and M-1 (*Populus canadensis*), on drought conditions, in order to indicate the potential of the studied clones for survival. Radical scavenging activity of poplar leaves was tested by ESR method using Fenton based hydroxyl radical production and 5,5-dimethyl-1-pyrroline-N-oxide as the hydroxyl radical traps. The content of proline and the activity of proline dehydrogenase (the enzyme which catalyzes degradation of proline), as well as quantity of malonyldialdehyde were determined. Cuttings of investigated clones were grown in Hoagland solution, and after 6 weeks, solutions of polyethylene glycol 6000 were applied with osmolarity: 100 mOsm and 200 mOsm, to induce a drought stress. Our results indicate that leaf extracts from all three clones exhibited high antioxidant activity. Prepared leaf phosphate buffer extracts of all three clones have shown high inhibitory effects on formation of hydroxyl radical. The best antioxidant activity has shown clone M-1, especially under 200 mOsm (strongly expressed draught conditions), and reduction was 27% compared with control. Significant increase of malonyldialdehyde content was observed only in clone M-1 under 200 mOsm, indicating that water deficit caused disruption of membrane integrity. The results demonstrate that proline accumulation was one of the main physiological plant response under drought, which had been mainly connected with the decrease in proline degradation (decrease of the activity of proline dehydrogenase). The accumulation of proline in all three clones indicated significant role of proline in drought tolerance.

**KEYWORDS:** drought, ESR spectroscopy, poplar, proline

Poster presentation 07 07 18

### CHARACTERIZATION OF DUTCH ELM DISEASE PATHOGEN POPULATIONS AFFECTING *ULMUS MINOR* IN CROATIA

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Dutch elm disease (DED) is a vascular wilt disease of elms caused by ascomycetes of the genus *Ophiostoma*. Two devastating pandemics of the disease have occurred in Europe and North America since the beginning of the 20<sup>th</sup> century. The first DED pandemic was caused by *O. ulmi*, which presently has been largely replaced by much more aggressive pathogen, *O. novo-ulmi*, causal agent of ongoing second DED pandemic. *Ophiostoma novo-ulmi* is separated into two distinct subspecies, ssp. *novo-ulmi* and ssp. *americana*, with weak prezygotic barrier to hybridization and emergence of ssp. *novo-ulmi*

x ssp. *americana* hybrids was reported in several European countries where ranges of two subspecies overlap. In Croatia, the disease was first recorded in Slavonia in 1929 and since then it is considered to be the most significant cause of the decline of elms in Croatian forests, especially affecting *Ulmus minor*. Recent investigation indicated that *O. novo-ulmi* is the only causal agent of DED in Croatia, while *O. ulmi* has probably completely disappeared. This study was conducted in order to determine distribution of *O. novo-ulmi* subspecies and estimate incidence of their hybrids in *U. minor* populations in Croatia. A total of 31 isolates of *O. novo-ulmi*, previously obtained from infected *U. minor* samples from three sites across Croatia (Nova Kapela, Đurđevac i Jastrebarsko), were analyzed by PCR-RFLP of *cerato-ulmin* (*cu*) and the colony type (*col1*) gene regions. Presence of both *O. novo-ulmi* subspecies was proven, but with significantly higher incidence of ssp. *novo-ulmi* at investigated sites. A twenty-one isolate was assigned to *O. novo-ulmi* ssp. *novo-ulmi* and only three isolates were assigned to ssp. *americana*. Seven isolates were shown to be subspecies hybrids. *O. novo-ulmi* ssp. *novo-ulmi* as well as hybrid isolates, were present at all three investigated sites, while *O. novo-ulmi* ssp. *americana* was detected only in Nova Kapela and Đurđevac. Results of this study represent significant contribution to understanding of the structure of DED pathogen populations affecting *U. minor* in Croatia.

**KEYWORDS:** *Ophiostoma novo-ulmi*, hybridization, subspecies hybrids, PCR-RFLP

Poster presentation 08 07 23

### MOLECULAR CHARACTERIZATION OF CHRYPHONECTRIA HYPOVIRUS 1 FROM SLOVENIA

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The pathogenic fungus *Cryphonectria parasitica* Murrill Barr has been responsible for the decline of European chestnut. This aggressive ascomycete causes chestnut blight, a serious disease that destroys chestnut trees by causing bark cankers that progressively enlarge, girdle and kill branches and trunks of infected trees. This disease in Europe is successfully controlled with naturally-occurring *Cryphonectria hypovirus 1* (CHV1), a double-stranded RNA (dsRNA) virus that reduces the virulence, sporulation and pigmentation of fungus and can therefore be used as a biocontrol agent of the chestnut blight. CHV1 was probably introduced together with its fungal host to Europe from Asia and then naturally spread throughout *C. parasitica* populations. In Slovenia, the disease

was first recorded in 1950 and has been reported to date in all investigated chestnut populations. *C. parasitica* in those chestnut populations has a high diversity of vegetative compatibility (vc) types that can limit the spread of CHV1. It is known that CHV1 easily spreads between *C. parasitica* strains of the same vc type, but between strains of different vc types it spreads less frequently. Despite a high diversity of vc types, CHV1 is widespread in Slovenian *C. parasitica* populations. In order to gain a better insight into the genetic diversity of CHV1, we have analysed CHV1 infected *C. parasitica* isolates from Slovenia. Molecular characterization of CHV1 included hypoviral dsRNA extraction, complementary DNA synthesis, PCR amplification and partial sequencing of CHV1 genome. The obtained nucleotide sequences were assembled and the number of nucleotide differences and genetic distance between them were determined. Phylogenetic analysis grouped CHV1 sequences from Slovenia to the Italian subtype of CHV1, the only subtype found so far in Slovenia. Among sequenced CHV1 isolates a large number of different haplotypes were detected which indicates a high genetic diversity of CHV1 in Slovenia. High genetic diversity is not a consequence of recombination events, but is probably the result of numerous point mutations.

**KEYWORDS:** biological control, chestnut blight, genetic diversity, hypovirulence

Poster presentation 09 07 13

### ROOT PHENOTYPING OF NS SUNFLOWER

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Sunflower breeding, in the past decades, had led to significant improvements in sunflower yield, pest resistance, and altered oil composition; however breeding is oriented towards analysis and improvement of above-ground parts, while root development has been significantly neglected. Future breeding efforts that would be aimed at modifying root traits can result in improved crops regarding stress-tolerance and ultimately increased yields by optimizing the capacity of the plant for soil exploration (water and nutrient acquisition). Development of novel phenotyping platforms for non-invasive root analysis facilitates characterization of root architecture and investigation of developmental dynamics and root growth. Up to our knowledge, this is the first report of sunflower root phenotyping using modern phenotyping platforms. In this preliminary study, one cultivated and one wild sunflower genotype were examined by use of the automated phenotyping platform, *GROWSCREEN-Rhizo*. Imaging of the rhizotron

grown plants had been performed twice per week and the following traits were quantified using the image processing software *GrowScreen-Root*: total root length, primary and lateral root length, rooting depth, root system width, and area covered by the root system. At the end of the experiment, fresh and dry shoot weight were measured. After harvest, sunflower roots were used for determining total root length and root diameter by utilizing the *WinRhizo* system. During plant development and imaging of the roots, it had been observed that cultivated sunflower developed faster comparing to the wild relative. Preliminary analysis of total root length after washing and the one obtained by imaging showed that approx. 1/3 of the whole root system is visible at the transparent surface of the rhizotrons.

**ACKNOWLEDGEMENTS:** This study was supported COST Action FA 1306: The quest for tolerant varieties -Phenotyping at plant and cellular level (STSM); by Provincial Secretariat for Higher Education and Science of Vojvodina, project 114-451-2126/2016-03; by Ministry of Education, Science and Technological Development of Republic of Serbia, project TR31025.

**KEYWORDS:** *H. annuus* L., root architecture, rhizotrons

Poster presentation 10 07 06

### COMBINED APPROACH FOR IDENTIFICATION OF PHENOTYPIC AND -OMICS MARKERS THAT COULD BE INCLUDED IN SUNFLOWER BREEDING PROGRAMS

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Diversity in plant genetic resources provides an opportunity for plant breeders to develop new and improved cultivars with altered quality traits, resistant to diseases and unfavourable environment. Novel approaches in genotyping and phenotyping enabled more efficient data collection for identification of quantitative characters and to explain the genetic basis of agriculturally important traits. The flip side of these new approaches is the risk of drowning in the massive amounts of data. That is why it is essential to develop proper approaches for data management and integrated analysis of differently collected data. Within the framework of ongoing projects, we have started to perform comparative phenotypic, metabolic and molecular analyses of 7 annual and 21 perennial wild sunflower (*Helianthus* spp.) species, as well as 19 genotypes of cultivated sunflower. The material consists of annual and perennial wild sunflower species (<http://www.nsseme.com/about/inc/oilcrops/wild.php>), interspecific hybrids, varieties, lines and hybrids, chosen from the IFVCNS collection, which is one of the largest sunflower germplasm collections. Data are

collected for 48 morphological and respective metabolic parameters. This is further complemented by molecular analyses for identification of molecular markers and QTLs correlated to parameters studied. The aim of this combined approach is to identify desirable traits and genotypes that could be further included in sunflower breeding programs for development of highly productive, stress resistant hybrids. A long-term goal is creation of ideotypes specific for certain agro-ecological conditions. Special attention is paid to the integration of phenotypic and -omics data in order to identify traits and markers of real practical value for the breeders, avoid massive collection of redundant data and render the process more efficient.

**ACKNOWLEDGEMENTS:** This study was supported by Ministry of Education, Science and Technological Development of Republic of Serbia, project TR31025, Provincial Secretariat for Higher Education and Science of Vojvodina, project No. 114-451-2126/2016-03, Serbian-German bilateral cooperation project No. 451-03-01732/2017-09/3 and COST Action CA 16212.

**KEYWORDS:** *Helianthus* sp., phenotyping, genotyping, breeding

Poster presentation 11 07 19

### DIVERSITY OF WHEAT GENOTYPES BASED ON MORPHOLOGICAL MARKERS

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Phenotypic, biochemical and molecular evaluations of wheat collections are of a great importance to increase the knowledge of genetic diversity as the basic prerequisite for crop improvement in different breeding programs. Large scale genotyping by molecular markers as well as phenotyping of agronomical important traits generated a lot of valuable information for wheat researchers during the last few decades. However, some morphological traits are almost forgotten and very rarely used in evaluating diversity of wheat germplasm. The aim of this study was to analyse the morphological diversity in a collection of 450 wheat accessions originating from all over the world. The genotypes were chosen from the wheat genetic collection of the Small Grains Department and sown at the experimental field of the Institute of Field and Vegetable Crops, location of Rimski Šančevi (45°20'N, 19°51'E). Five morphological traits were analysed and used as markers for distinctness of wheat genotypes: auricle colour (AC), coleoptile colour (CC), leaf colour (LC), colour at tillering time (CTT) and grain colour (GC). The Shannon diversity index (H) was estimated as a measure of morphological diversity. The results have shown that the most of the genotypes had white auricle colour (87%), white coleoptile colour (81%), dark green colour at tillering time (96%), green leaf colour (61%) and light red grain colour (43%). The average value for

the Shannon diversity index was 0,77 indicating high level of morphological diversity in the collection. The lowest diversity (0,07) was found for CCT and the highest individual descriptor diversity was found for GC (0,68). The results indicated that morphological characterization with higher number of analysed traits can be used to improve description of wheat germplasm in addition to molecular and agronomical evaluations.

**KEYWORDS:** variability, morphological characters, Shannon index, *Triticum* sp.

Poster presentation 12 07 20

### ENVIRONMENTAL MODELING OF INTERACTION VARIANCE FOR GRAIN YIELD OF MIDDLE-LATE MAIZE HYBRIDS

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The phenomenon of genotype by environment interaction (GEI) represents permanent interest for breeders and biometricians along with practical and theoretical aspects. We investigated GEI for grain yield of middle-late maize (*Zea mays* L.) hybrids from the official variety trial network by the Department of Protection and Recognition of Varieties of Republic Serbia that includes 25 experimental maize hybrids in the year of 2004, and 15 in the year of 2005, on eight sites over two years, using factorial regression model that captures additional environmental variables - maximum temperature (mxt), minimum temperature (mnt), mean temperature (mt), precipitation (pr), relative humidity (rh), insolation hours (in) and environmental index (EI). Environmental variables in the best of the tested models explained 76% and 76.2% of the GEI variation for two consecutive years, respectively. Factorial regression combined with stepwise procedure revealed the 5-variable model that includes variables pr7 (explaining 29.1% of the variation), EI (19.1%), mnt9 (14.5%), mxt4 (13.3%), in 2004 and the two-variable model: mnt6 (60.2%), mt9 (16.0%) in 2005 as the most explanatory models in the region of Vojvodina for these two consecutive years. These results provide a solid base for further research in GEI and stability analysis, and are a useful tool in characterizing the sub-regions of maize growing area and extending the existing results to new sites.

**KEYWORDS:** genotype by environment interaction, maize, yield, environmental variables

Poster presentation 13 07 24

### EVALUATION OF THE BALKAN PHASEOLUS COCCINEUS L. GENETIC RESOURCES

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In the last decade several thousand accessions were collected in different parts of Europe and are stored in national gene banks; at present the *Phaseolus* Database as part of the EURISCO Web catalogue contains over 46000 records. Main objective of the study (ECPGR\_SMARTLEG project) was proper phenotypic and genetic evaluation of European *Phaseolus* accessions with emphasis to *Phaseolus coccineus* germplasm providing new data to EURISCO and AEGIS. During the project, we provided three different types of datasets including morpho-agronomic traits of plants, morphometric seed characteristics and genetic profiles of *P. coccineus* accessions from eight European geographic origins; Slovenia, Romania, Bosnia and Herzegovina, Serbia, Italy, Slovakia, Macedonia and Austria. Before sowing, the morphometric characterization of *P. coccineus* seeds was performed according to adopted Community Plant Variety Office-Technical Protocol and Phaselieu/AIS descriptors observing 14 different seed characteristics. The morphological evaluations were performed upon adjusted descriptors for *Phaseolus* (ECPGR\_PhaseChar from Austria) under field conditions in Slovenia, Romania, Serbia, Macedonia and Bosnia and Herzegovina. A set of 12 already proven and reliably cross-species amplified nSSR markers among *Ph. vulgaris* and *P. coccineus* genomes was applied to evaluate genetic structure of *P. coccineus* genotypes. Morpho-agronomic evaluation shows differentiation between *P. coccineus* accessions even for standard varieties under geographically distinct field conditions. Global principal component analysis extracted four components which cumulatively explains 74,6 % of morphometric variability of *P. coccineus* seeds. Analysis of molecular variability on the basis of allelic patterns reflects 3 % of the molecular variability among *P. coccineus* germplasm from eight different geographic origins which means that 97 % of germplasm is common to all accessions ( $p > 0.01$ ). Regarding to genetic structure of the *P. coccineus* germplasm from eight geographically distinct collections, three genetic clusters were formatted; the average genetic distance between genotypes in each cluster varies between 0.592 and 0.816. Overall results indicate common geo-

graphic origin of *P. coccineus* accessions from different collections within the Balkan.

**KEYWORDS:** morpho-agronomic traits, morphometric characterisation, genetic structure, ECPGR-SMARTLEG project, EURISCO

Poster presentation 14 07 05

### WILD FRUIT CROPS: PRESERVATIONS AND UTILIZATION

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Due to specific phyto-geographic relations, specific climatic, orographic and edaphic conditions forest phytocoenoses in Vojvodina (North Serbia) are very diverse. Significant and completely unused parts of this biodiversity are autochthonous wild fruit species. Due to the unique variability of the species, the Balkan peninsula can be considered as the secondary center of genetic diversity of several species and genera, such as: *Malus × domestica*, *M. sylvestris*, *Pyrus communis*, *Prunus cerasifera*, *P. mahaleb*, *P. fruticosa*, *P. tenella*, *Morus spp.*, *Corylus colurna*, *Castanea sativa*, *Fragaria vesca*, *Olea europea*, *Cornus mas*, *Sambucus nigra*, *Sorbus spp.*, *Rosa spp.* These wild and underutilized species produce fruits of unique quality and high nutritional value, which are used in human nutrition, food and pharmaceutical industries. Elderberry (*Sambucus spp.* L.), one of the most common and widespread species, can be considered as a source of natural antioxidants with promoting effect on human health. Eight divergent genotypes of elderberry were selected from natural populations of Central Serbia and Vojvodina with high antioxidant capacity and high content of vitamin C. Within genus *Morus* three species were analyzed and underwent selection. Selections of *Morus nigra* and *Morus rubra* were distinguished by high content of vitamin C. Genotypes with dark colored fruits contained high amount of anthocyanins and total phenols. Cornelian cherry (*Cornus mas* L.) holds great biological and economic potential, and it is rich source of material for further breeding. Within natural populations in whole Serbia high diversity considering fruit weight was observed. Five genotypes with large fruits and high soluble solids content were selected. *Sorbus domestica* exhibited high diversity level in Vojvodina, and on account of that selection of superior genotypes is currently in progress.

**KEYWORDS:** biodiversity, variability, selection, Balkan peninsula

Poster presentation 15 07 17

### EXPLORING AND GATHERING THE SERBIAN AND CZECH CROP WILD RELATIVES FOR INCREASING CROP DIVERSITY IN AGRICULTURE

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Conservation of plant genetic resources for food and agriculture is a very important task from the strategic and economic points of view almost in the whole world. Collecting expeditions and gathering of crop wild relatives, such as meadow (field and vegetable crops), aromatic, medicinal plants and some threatened species, are important parts of plant genetic resources activities which enrich gene bank storage composition. Common collecting missions have been organized annually in both countries since 2011. Seven of them took place in Serbia and seven in the Czech Republic. In total, 228 localities have been visited and 1202 seed samples of crop wild relatives have been collected. In the Czech Republic it was 107 localities and 659 seed samples, while in Serbia it was 121 localities and 543 seed samples. The numbers present large source of biodiversity, which can be utilized in breeding and within *in situ* conservation activities. All the samples are stored in the working collections of both institutes, after regeneration and multiplication process the seeds are stored in both national gene banks. These collecting missions provided new information concerning the distribution of crop wild relatives and traditional farming practices still present in some regions, especially in Serbia. Our preliminary results indicated that the wild seed lots are an important source of valuable genetic variability for utilization in breeding programs. In the paper detailed survey of collected material and its utilization is presented.

**KEYWORDS:** genetic resources, collecting missions, Serbia, Czech Republic, gene banks, conservation, evaluation

Poster presentation 16 07 08

### OXIDATIVE STRESS IN SOYBEAN SEEDLINGS INOCULATED WITH *BACILLUS SUBTILIS* FOLLOWED BY MITES (*TETRANYCHUS URTICAE*) ATTACK

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The control of crop pests is one of the main problems in agriculture. Use of chemical pesticides and fertilizers cause serious environmental pollution and health problems. Due to that, modern agriculture is facing new challenges where microbial biocontrol agents are being integrated to achieve higher crop yields with minimizing environmental pollution. Growth-promoting rhizobacteria *Bacillus subtilis* has been documented as biological control agent and elicitor of induced systemic resistance (ISR). *Tetranychus urticae*, well known as mite, is an occasional pest of soybean that cause biotic stress. In response to pest invasion, plants produce reactive oxygen species (ROS) which are highly reactive and can seriously damage vital biomolecules. Plants developed antioxidant protective mechanism against oxidative stress. Phenolic antioxidant molecules have radical-scavenging capacity and play an important role against ROS. This study was conducted in order to assess the effect of inoculation of soybean (*Glycine max* L.) seeds with *B. subtilis*, followed by mites (*T. urticae*) exposure, on the content of total phenolics and tannins. In addition, radical scavenging activity of extracts was investigated using 2,2-diphenyl-1-picrylhydrazyl (DPPH) assay. The total phenolic and tannins content was determined according to the Folin-Ciocalteu method and DPPH assay was done according to the method of Lee et al. (1998). The experimental design included inoculated and non-inoculated soybean seedlings – with, and without exposure to mites. It has been shown that *B. subtilis* does not cause oxidative stress in plants and could be used in practice. On the other hand, mites attack triggered oxidative stress in plant leaves and roots and, thus, significant accumulation of total phenolics and tannins. Inoculated soybean seedlings treated with mites had lower content of phenolics and tannins compared to non-inoculated group treated with mites which points to protective role of *B. subtilis*. The highest scavenging activity was detected in inoculated group (DPPH: 45,46±0,51 mg trolox/gdw), and the lowest activity was in inoculated group treated with mites (DPPH: 27,12±0,90 mg trolox/gdw). The obtained results support the idea that inoculation of soybean seeds with *B. subtilis* improve resistance of soybean seedlings against mites attack. Therefore, *B. subtilis* potentially may be utilized as biopesticides.

**KEYWORDS:** biotic stress, antioxidants, antioxidant potential, *Glycine max* L., *Bacillus subtilis*, *Tetranychus urticae*

Poster presentation 17 07 09

### STORAGE POSSIBILITY OF THE PRIMED SOYBEAN SEED (*GLYCINE MAX* (L.) MERR.)

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In the technology production, various seed enhancement procedures are often applied to improve the quality of the seed, in particular germination and other properties of the seed after the treatment. Seed priming is the method used by the Old Greeks. Today it is a cheap and simple presowing measure that is primarily used in developing countries. However, the biggest problem in applying this measure is the length of storage of the primed seed. Storage possibility of the primed soybean seed was determined by a temperature of 25 °C during the period of 90 days. The soybean seed was primed with KNO<sub>3</sub> (1%), ASA (100mg l<sup>-1</sup>) and KCl (1%) solutions, and then stored in paper bags, and its quality is tested every 15 days. The results showed that the reduction in the quality of the primed seed is considerably faster than the no-primed. Primed soybean seed can be stored at a temperature of 25 °C during 60 days after immersion, and after that period the significant reduction in its quality can occur. After 75 days of immersion germination energy was reduced by 60.33% and germination by 9.33%, while after 90 days the germination energy reduction was 68.33% and germination by 65%. One of the causes for reducing seed quality is an increase in MDA content, especially after 75 and 90 days of storage. Also, the free proline concentration has been reduced while the content of vitamin C increased after 15 days primarily in seeds immersed in ASA and KCl solutions, and after 45 days its content was reduced.

**KEYWORDS:** seed priming, seed storage, soybean

Poster presentation 18 07 15

### ANTIFUNGAL ACTIVITIES OF IONIC LIQUIDS AGAINST PHYTOPATHOGENIC *ALTERNARIA* SPP. STRAINS

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A modern sustainable approach in agricultural practice would be a replacement of synthetic and toxic substances in the treatment of crop with benign and environmentally friendly compounds, by using principles of green chemistry. One of the possible directions could be using of ionic liquids (ILs), salts with melting points below 100 °C, which are well-known for manifesting antimicrobial activity. ILs can be used in the development of novel sources of antimicrobial agents such as antiseptics, biocides and antifungal agents. Furthermore, ILs have already been reported as alternative 'green' solvents for a wide range of reactions and technological processes. Considering the importance of research in the area of bioactivity of ILs for discovery of new green ILs for different purposes, the aim of this study was to examine antifungal activity of nine newly synthesized ILs against phytopathogenic *Alternaria* strains isolated from rice (*A. padwickii*), carrot (*A. dauci*) and linseed (*A. linicola*). Antifungal effect was estimated by micro-plate microdilution method for establishing minimum inhibitory (MIC) and minimum fungicidal concentration (MFC). All ionic liquids showed strain specific but good antifungal activity on *Alternaria* strains with MIC and MFC detected at the range from 9.23 mg/ml to 75.89 mg/ml. Only in case of the 1-(4-hydroxy-2-oxy)butyl -3-methylimidazolium chloride [OHC2OC2mIm][Cl], no antifungal effect on *A. dauci* were observed. Comparing to *A. padwickii* and *A. dauci*, *A. linicola* showed higher sensitivity to all tested ILs. The obtained results indicate the possibility of usage of ILs in biocontrol of plant diseases, representing their application in crop protection. However, further research is necessary in order to examine their toxicity and biodegradability in the environment.

**KEYWORDS:** agriculture, *Alternaria*, antifungal activity, green chemistry, microdilution method, phytopathogenic fungi

Poster presentation 19 07 14

### BACILLUS STRAINS AS POTENTIAL AGENTS FOR THE BIOCONTROL OF PHYTOPATHOGENIC FUNGI *ALTERNARIA* SPP.

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Plant diseases caused by fungal pathogens can lead to major economic losses in agriculture. The genus *Alternaria* includes saprobic, endophytic and pathogenic species that may cause various plant diseases such as blight disease. Symptoms of *Alternaria* blight include the presence of irregular, often circular, brown to dark brown coloured leaf spots. *Alternaria* spp. also cause collar rots, stem lesions, tuber and fruit rots of their respective hosts. In addition, bacteria are one of the most frequently biocontrol agents used to protect plants from diseases. *Bacillus* species control disease through a variety of mechanisms. Bacterial antagonists might act as inhibitors of growth, development and reproduction of pathogen, or as inducers of host resistance in plant. The objective of this study was to examine *in vitro* antifungal activity of ten antagonistic *Bacillus* strains from collection of Department for Microbiology of Institute of Field and Vegetable Crops, Novi Sad. Strains were originally isolated from the soil samples collected from several localities of Vojvodina Province. Antifungal activity of *Bacillus* strains against *Alternaria padwickii*, *A. dauci* and *A. linicola*, obtained from rice, carrot and linseed, respectively was tested using a dual plate assay. The results confirmed that all tested *Bacillus* strains showed antifungal activity against *Alternaria* spp. The highest antagonistic activity was exhibited by *B. pumilus* B11 (45.71% to 51.25%), *B. subtilis* B13 (45.71% to 49.37%) and *B. subtilis* B32 (45.71% to 52.08%), while *B. pumilus* B23 had the weakest antifungal activity. *B. safensis* B2 (35.24% to 48.33%), *B. pumilus* B21 (40.95% to 48.75%) and *B. pumilus* B22 (33.50% to 49.17%) also demonstrated good antifungal potential. *A. dauci* was the most sensitive fungus, while the most resistant was *A. linicola*. Obtained results indicate the possibility of usage of the most effective *Bacillus* strains as potential biocontrol agents of plant diseases caused by *Alternaria* spp.

**KEYWORDS:** agriculture, *Alternaria*, antifungal activity, *Bacillus*, biocontrol, phytopathogenic fungi

Poster presentation 20 07 02

## ALIEN PLANTS IN WEED FLORA OF ORGANIC CROPS IN PANNONIAN PART OF SERBIA

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On the basis of long-term (2008-2015) floristic studies of weeds in organic crops on the largest certified farms for organic plant production in Pannonian part of Serbia, was analyzed the presence of alien weed species while special emphasis was placed on the time of the introduction, their life history, origin and degree of invasiveness. During the studied eight-year period, a total of 85 species were found in different organic crops of which 52 species (61%) are alien plant, classified in 21 families, the most numerous are the families Asteraceae (14 species; 27%) and Poaceae (8 species; 15%). Of the total number of alien plants, 43 species (83%) belong to archaeophytes, of which the most common are naturalized non-invasive species (37; 71%), while only one (2%) is casual species and 5 (10%) are invasive. Neophytes account for 9 species (17%), most of which are invasive (5; 10%), followed by naturalized non-invasive species (3; 65%) and only one (2%) casual species. The analysis of certain categories of alien species shows that out of 14 species from the family Asteraceae, 10 are archeophytes, of which 9 are naturalized and one is invasive, while there are 4 neophytes, and all of them are invasive. This family was followed by Poaceae, which included 7 archeophytes, 5 naturalized and 2 invasive species, and only one neophyte from casual species. The remaining 19 families were represented with a small number of species or only one representative, of which the study focuses on three invasive species, including one neophyte from Amaranaceae and two archeophytes from Portulacaceae and Apiaceae. Considering the life history, these agroecosystems are dominated by annual plants (41 species; 79%). Perennial species are less common (10; 19%), while there is only one biennial species (2%). With regard to their origin, the most of the studied alien weed species originate from the Mediterranean (34 species; 66%), followed by America (7 species; 13%), a wider area of Europe (6 species; 12%) and Africa (1 species; 2%), while 4 species (8%) are from the category of anecophytes, the origin of which is unknown.

**KEYWORDS:** weeds, archaeophytes, neophytes, invasive plants

Poster presentation 21 07 25

## WEED FLORA COMPOSITION IN ORGANIC CULTIVATIONS OF AROMATIC PLANTS

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Weeds are usually seen as constraints on crop production, however, they are major components of the agroecosystems biodiversity and are involved in many useful ecosystem services. It is known that many aromatic plants have allelopathic activity. The possible effects of this activity on the composition of the weed flora in cultivations of aromatic plants has not been sufficiently investigated. Our aim was: (a) to identify the weed flora in cultivations of different aromatic plants, and (b) examine the diversity in weed flora composition according to the cultivated species. We studied in total 14 organic cultivations including oregano (*Origanum vulgare* subsp. *hirtum*), mountain tea (*Sideritis scardica*, *S. clandestina* subsp. *peloponnesiaca*), thyme (*Thymus vulgaris*, *Th. sibthorpii*), sage (*Salvia officinalis*) and lemon balm (*Melissa officinalis*), located in five farms near Thessaloniki (N Greece). We used a quadrat-based sampling method (448 plots of 0.5 m<sup>2</sup>, i.e. 32 in each cultivation) over a 4-month period. In each plot we recorded all the present weed taxa, the coverage of each species and the total weed coverage. A total of 72 taxa belonging to 26 families were found. These are mostly therophytes (70% of the taxa) with Eurasiatic (42%) or Mediterranean (30%) distribution. The most frequently occurring taxa which also have high coverage values are *Digitaria sanguinalis*, *Polygonum aviculare*, *Convolvulus arvensis*, *Portulaca oleracea* and *Lolium rigidum*. The oregano cultivations had the lowest average weed coverage. Our results showed heterogeneity in the composition of the weed flora among the cultivations of the different aromatic plant species, even at the same farm. However, the weed flora composition was more similar between the different aromatic species cultivated at the same farm, when compared to the weed flora found for a given species cultivated at other farms. Although the cultivated species may influence the composition of the weed flora, it appears that environmental factors or cultivation practices may have a greater impact between different regions.

**KEYWORDS:** weed flora, cultivations of aromatic plants, oregano, mountain tea, thyme, sage

Poster presentation 22 07 26

## A CASE STUDY OF ECOLOGICAL WEED SPECIES ABUNDANCE IN IMO STATE UNIVERSITY, OWERRI, IMO STATE, NIGERIA

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One of the more utilized methods for the analysis of weed communities in agricultural systems is the phytosociological study. Phytosociological studies of weeds are necessary for understanding the relationship between crops and their weed flora and may be useful, as a tool for developing a sustainable long-term weed management strategy. Study was conducted to assess the phytosociological studies of weeds on twelve sites in Imo State University Owerri, Imo State. The study was conducted during the 2016 wet season. A total of 21 weed species were identified during the investigation. The results obtained indicated that *Ipomea tribola*, *Talinum triangulare*, and *Sida garckeana* were the most densely populated in all the sites. Phytosociological investigation showed an *Ipomea-Talinum-Sida* plant community.

**KEYWORDS:** weed species, weed abundance

Poster presentation 23 07 04

## INFLUENCE OF SOME HERBICIDES AND THEIR MIXTURES WITH GROWTH REGULATOR AND FOLIAR FERTILIZER ON THE PRIMARY ROOT LENGTH OF COTTON SEEDS (*GOSSYPIUM HIRSUTUM* L.)

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The trial was carried out during 2013-2015, with two cotton cultivars – Helius and Darmi (*Gossypium hirsutum* L.). The effects of herbicides Goal 2 E (oxifluorfen), Linuron45 SC (linuron), Wing P (pendimethalin + dimethenamid), Merlin 750 WG (izoxaflutole), Bazagran 480 SL (bentazone) on the primary root length were studied. These herbicides were used alone or in combinations with the growth regulator Amalgerol premium or with the foliar fertilizer Lactofol O during the budding stage of cotton. From the viewpoint of cotton growing technology, technologically the most valuable are combinations of all herbicides with Lactofol O, which are followed by Linuron450 SC + Amalgerol, Wing-P + Amalgerol, Merlin 750 WG + Amalgerol, Bazagran480 SL + Amalgerol and sole use of herbicide Goal2 E on cultivar Helius. Techno-

logically the most valuable are herbicides Goal 2 E, Wing-P and Bazagran480 SL and tank mixtures Wing-P + Amalgerol, Merlin 750 WG + Amalgerol, Wing-P + Lactofol O, Merlin 750 WG + Lactofol O and Bazagran480 SL + Lactofol O on cultivar Darmi. These variants combine high biggest primary root length and high stability of this index during the different years. The sole use of the herbicide Linuron45 SC has low assessment and should be avoided.

**KEYWORDS:** cotton, herbicides, foliar fertilizer, growth regulator, primary root length

Poster presentation 24 07 03

## INFLUENCE OF FOLIAR ANTIBROADLEAVED HERBICIDES ON THE PRIMARY GERM WEIGHT OF COTTON SEEDS (*GOSSYPIUM HIRSUTUM* L.)

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The trial was carried out during 2013-2015, with twelve cotton cultivars (*Gossypium hirsutum* L.). Influence of herbicides Bazagran 480 SL (bentazone), Pulsar 40 (imazamox) and Express 50 SX (tribenuron-methyl) was studied. These herbicides were used during the budding stage of cotton. From the viewpoint of cotton growing technology, technologically the most valuable are cultivars Helius, Trakia, Viki, Filipopolis, Boyana, Avangard, Natalia, Darmi, Dorina and Nelina, by foliar treatment with herbicide Bazagran 480 SL. Technologically the most valuable are cultivars Chirpan-539, Helius, Viki, Boyana and Natalia by foliar treatment with herbicide Pulsar 40. Technologically the most valuable are cultivars Helius, Trakia, Viki and Nelina by foliar treatment with herbicide Express 50 SX. These variants combine high primary germ weight and high stability of this index during the different years.

**KEYWORDS:** cotton, herbicides, foliar treatment, cultivars, primary germ weight

## Session 8. Botanical Collections and History



Balkan  
Botanical  
Congress

Introductory lecture 01 08 12  
**BALKAN HERBARIA: DO WE HAVE TO  
WORRY ABOUT THEM?**

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In the area of Balkan Peninsula, according to Index herbariorum there are 57 herbarium collections with around 5,5 million herbarium sheets of vascular plants. It may be an impressive figure, but in fact it is comparable to the size of only one big Vienna herbarium (which, among others, holds also important collections from the Balkans). Despite the fact that serious work in several fields of botany is not possible without studying, sampling or depositing herbarium material, existence and needs of maintenance of herbarium collections are often forgotten. Our aim was to get a better overview of the current status of Balkan herbaria so all of them were invited to fill-in a survey. The first problem, that had been expected, was old contact details, which caused only about 1/3 immediate response rate. Already the preliminary results showed that our herbaria are mostly part of different institutions (universities, museums, research institutes), herbarium collections are mostly not protected by national legislation, work of herbaria is mostly financed by institutional budgets, to a lesser extent from projects. In average there is about 1/4 to 1/3 of herbarium material not labeled and waiting for inclusion in the collection, and fortunately our herbaria have in average about 1/3 of room potential for expansion. In average herbaria have only one person employed (but some of them zero). The most frequent pest problems are caused by moulds, less often with some booklice or bugs, and the most commonly used pest control measure is regular deep-freezing of herbarium material. Unfortunately air-conditioning system is mostly not used at all. Herbarium material exchange frequency is low, in average less than 10 packages per herbarium per year and so is the number of foreign visitors. About half of collections are databased and about a third digitalized, but data are often not publicly accessible. General impression of the situation in particular herbarium is, that financial situation and availability of staff is very bad to bad, technical conditions mostly sufficient, but awareness of the problems on higher decision-maker levels (directors, government etc.) also mostly bad. So obviously we will all have to work harder to raise the awareness of importance of herbarium collections in public and maybe even more particularly among colleagues.

**KEYWORDS:** herbarium collections, museum, research institutes, herbarium pest

Oral presentation 02 08 09  
**ANDREAS WOLNY'S HERBARIUM, TREASURE  
OF THE SREMSKI KARLOVCI GRAMMAR  
SCHOOL: 1797-2017 STATE-OF-THE-ART**

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Andreas Wolny's herbarium collection, stored in Karlovci grammar school, is the oldest collection in Serbia. It is established at the end of the 18 century, by Andreas Wolny, who taught botany among other subjects in school. Wolny was a respected botanist at the time, and had fruitful cooperation with Pál Kitaibel. After leaving Sremski Karlovci, he leaves in the legacy representative herbarium collection. In the period after him, professors and pupils continued his work on herbarium enlargement. During 19 century, school reform has led to the discontinuation of activity in herbarium. In the mid of the 20 century, the entire collection was placed under state protection as important cultural, historical and scientific heritage. After that, only few visits to herbarium by specialists are documented. However, authority kept herbarium under protection, with only changes of storage room. Only data from revision study that was performed for the legal protection protocol was available to scientific and professional public until recently. Society was mainly without any knowledge about collection, as well as school employees and pupils. In previous five years, initiative to re-open this herbarium for scientist and general public as well was raised. First task was access and insight in collection, to define current state-of-the-art. For that, storage cabinet is inspected, and part of material is documented. Process and specific exsiccata are digitized. It is confirmed presence of: two centuries created by Andreas Wolny, herbarium collected by Professor Josif Pančić, collection that contains material obtained by exchange from foreign institutions and botanists during 19 century and the collection of professors and pupils of the school. The second task was to find all relevant documents, scientific papers and publications that would give us a closer look at the development of botany and herbarium. For that reason, one part of school library was inspected, and documents in Matica srpska library were consulted as well. Currently, only one part of collection is uncovered, and known data are only complemented. Important findings are book and articles press at the beginning of 19 century and special herbarium collections.

**KEYWORDS:** botany, history, collection, museum, Andreas Wolny

Oral presentation 03 08 04

### DIGITISING HISTORICAL BOTANICAL COLLECTIONS FROM A PHYTODIVERSITY HOT SPOT

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Taxonomy provides the foundation to understand biodiversity and such derived biological disciplines as biogeography, ecology and evolution. Specimens are the faithful custodians of taxon identity, contributing the fundamental proofs for solving systematic issues. The preservation of scientific collections and their availability for study over time has been an everlasting concern of scientists. The Thessaloniki Aristotle University (TAU) Herbarium currently holds about 50,000 plant specimens collected mainly from Greece from the beginning of 19<sup>th</sup> century. It comprises historical specimens collected up to the first half of the 20<sup>th</sup> century (before World War II) by important botanists such as Th. von Heldreich, who described seven new plant genera and over 700 species, V. Tuntas, Ch. Leonis, and D. Zaganiaris. The latter, in his "Herbarium Macedonicum", recorded 4,000 plant taxa, collected from the northern part of the country. Our effort is to digitise the TAU Herbarium, facing the challenges and getting ahead the limitations related to the effective mobilization and sharing of collections information from small herbaria and with limited resources. Digitisation will serve not only taxonomic, biodiversity conservation, and educational purposes, but also will reveal the historical and socio-political aspects, under which the immense plant diversity of Greece was assessed. We have set up a pre-digitisation process which includes nomenclature checking and annotation (where needed), imaging, data transcription, and incorporation of all the information linked to the specimens in a database confronting with modern biodiversity information management requirements, such as the Darwin Core biodiversity metadata Standard, in order to be made publicly accessible and interoperable. Priority will be given to the historical collections, since the evaluation and digitisation of the old specimens may provide valuable information about the past species distribution and habitats. This work aims to trigger a discussion around the challenges of the digitization and the importance of small Herbaria.

**ACKNOWLEDGEMENTS:** The TAU equipment was funded by the Greek Ministry of Education, Research and Religious affairs through the Rectorate and the Deanship of the Faculty of Sciences of AUTh for the educational purposes of the Postgraduate Studies Program "Conservation of Biodiversity and Sustainable Exploitation of Native Plants", School of Biology

**KEYWORDS:** Greece, TAU Herbarium, historical collections, digitalisation

Oral presentation 04 08 14

### ORCHID DIVERSITY WITHIN HERBARIUM CROATICUM SENSU STRICTO AND HERBARIUM IVO AND MARIJA HORVAT COLLECTIONS

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Following the recent trend in herbaria, digitisation of collections within *Herbarium Croaticum* (ZA) and *Herbarium Ivo and Marija Horvat* (ZAHO) has initiated in 2015. With over 200.000 herbarium specimens, ZA Collection is the oldest and the largest herbarium collection in Croatia, while the ZAHO Collection stores as many as 78.000 specimens. Digitised collections allow public access to such information via herbarium-based databases (virtual herbaria), which makes them more broadly useful and improves scientific research. In our study, a large collection of orchids was chosen for digitisation. Our aims were to digitise the collection of orchids and analyse the related herbarium sheets to provide the taxonomical, spatial and temporal data and, finally, estimate the average time required to digitise one herbarium sheet. Prior to digitising, we verified the taxonomy of our specimens, revealed synonyms and specified the hybrids. Plant material was cleaned, dusted, translocated and mounted with pH neutral adhesive tape to new format of herbarium sheets adequate for scanning, on which the herbarium labels were also glued. Herbarium sheets were scanned using the inversed Epson Expression 11000XL Pro A3 scanner, to produce image files with .tiff extension, with resolution of 300 dpi. Finally, high resolution images, together with the metadata of digital herbarium, were uploaded and published in Flora Croatica Database, as well as in ZA & ZAHO Virtual Herbarium. Analysis revealed that the collection of orchids within *Herbarium Croaticum sensu stricto* contains 1373 herbarium sheets, belonging to 92 taxa, including 15 subspecies and 4 hybrids. Two orchid holotypes and two isotypes have been found. Within ZAHO Collection 553 sheets were found, belonging to 47 taxa. The majority of both collections originates from Croatia and neighboring countries (Slovenia, Serbia, Bosnia and Herzegovina, Italy, FYROM, Montenegro, Poland, Switzerland and Ukraine). The oldest sheet in ZA dates back to 1833, and in ZAHO back to 1918, while the average age of the ZA Collection is 113 years and of the ZAHO is 81. The main collectors in ZA are Ljudevit Rossi, Dragutin Hirc and Ambroz Haračić, while in ZAHO predominates Ivo Horvat.

**KEYWORDS:** Croatia, digitisation, herbarium, orchids

Poster presentation 05 08 07

### VIRTUAL HERBARIUM OF ZA AND ZAHO COLLECTIONS (ZAGREB, CROATIA)

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Natural history collections contain an invaluable source of information in a variety of biological research. In addition to the specimens themselves, collections contain detailed supplementary data such as the name of the species, the collector, and the collection time and location. For scientists, the access to relevant herbarium material is often challenging due to the very broad or relatively narrow distribution of certain taxa. In order to increase the availability of collections and at the same time ensure its permanent protection, especially of historically significant material, the general trend is that the collections are digitized and presented in publicly available databases. The digitization of herbarium collections involves the process of storing data and images in digital form which enables easier dissemination of data and access to collections. The use of digital images in publicly available databases contributes to the preservation of herbarium collections, as it reduces the need for direct handling, borrowing and transportation of copies, while at the same time high-quality digital images make it possible to observe most morphological details. Within the Division of Botany of the Department of Biology, Faculty of Science, University of Zagreb there are two officially registered herbarium collections: *Herbarium Croaticum* (ZA), and *Ivo and Marija Horvat herbarium* (ZAHO). These are the two oldest and largest collections in Croatia. The first collection contains herbarium specimens collected by various researchers during the past 200 years, while the second collection is the fruit of lifetime labour of professor Ivo Horvat and his wife Marija. The digitalization of these two collections started in 2015 and is continuously done through the Flora Croatica Database. In order to provide easy on-line access to the digitalised material the Virtual herbarium was developed as a part of *Herbarium Croaticum* web interface - <http://herbariumcroaticum.biol.pmf.hr/>. A user-friendly search engine was created enabling the search of herbarium specimens across several most relevant queries. The easy access to digitized herbarium specimens is expected to enhance their use in scientific research as well as to broaden the use of botanical collections in citizen science.

**KEYWORDS:** virtual herbarium, ZA, ZAHO, Croatia

Poster presentation 06 08 01

### "LOST WORLDS OF ARCHAIC GARDENS": EXHIBITION IN THE BOTANICAL GARDEN OF THE FACULTY OF SCIENCE, UNIVERSITY OF ZAGREB

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In 2018, Zagreb Botanical Garden of the Faculty of Science presents a new popular scientific exhibition created by the Garden curators, in cooperation with the Faculty's botanists, palaeontologists, geographers and chemists. The exhibition, named "Lost Worlds of Archaic Gardens" ('Izgubljeni svjetovi - pradávnii vrtovi devona, karbona i krede') depicts the evolution of the Plant Kingdom through the imaginary "gardens" of three well-known periods in the geological time scale: early Devonian (Palaeozoic), late Carboniferous (Palaeozoic) and mid-Cretaceous (Mesozoic). Through three dioramas - fictional garden beds from the prehistoric times - the visitors are able to learn how and when the plants conquered land and through hundreds of millions of years finally brought us to the world we know today. In this manner, the plant collections of the Garden's greenhouses and arboretum are presented to the visitors in a new way, revealing their age and origin according to the latest scientific research. The exhibition, originally intended for schoolchildren and students primarily, is also of great interest to the widest Garden audience.

**KEYWORDS:** scientific exhibition, Garden collections, prehistoric gardens, plant evolution, Zagreb Botanical Garden

Poster presentation 07 08 15

### COLLECTIONS OF TROPICAL AND SUBTROPICAL PLANTS IN THE BOTANICAL GARDEN, BULGARIAN ACADEMY OF SCIENCES

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The collections of the Botanical Garden, BAS in Sofia have more than 100 years history. The position of the Garden has been moved three times during this period and the construction and development at the present place is not completed. The 'glasshouse' collections of tropical and subtropical plants are the richest ones, with about 3100-3200 taxa. They are situated in 12 departments with an area 2300 m<sup>2</sup>. A general review of the history of collection, development and documentation, taxonomic structure and uses is presented. The



species are about 2500 from 170 plant families. The richest are the collections of Cactaceae, Orchidaceae, Crassulaceae, Araceae, Mesembryanthemaceae, Geraniaceae, Gesneriaceae, Bromeliaceae, Begoniaceae, etc. Many species with a conservation status are presented, especially in Cycad collection (12 species, 7 genera, 2 families), Cactaceae and Orchidaceae collections. Cultivars constitute about 20% of all taxa. The richest in cultivars family's collections are those of Geraniaceae, Gesneriaceae, Moraceae. Historical part includes century old individuals of cycads, palms and strelitias as well as almost 70 orchid and succulent taxa, vegetative propagated during many decades. The larger part of the items are received by means of the international seed exchange between the botanical gardens, followed by the samples obtained during personal visits in other gardens. During the last 2 decades increases the number of accessions that are donated by visitors. The glass-houses have been opened to the public since 1998 and visited by 8-15000 visitors annually. Students of five universities have practical classes there. Some exhibitions and events are organized every year in the Garden. Among them are now traditional exhibitions of azaleas and orchids, such for systematic, ecological or economic group of plants (ferns, begonias, succulents, pelargoniums, etc.).

**KEYWORDS:** Botanical garden – BAS, Sofia, collections of tropical and subtropical plants

Poster presentation 08 08 05

### HERBARIUM OF VASCULAR PLANTS COLLECTION OF THE UNIVERSITY OF PATRAS (GREECE)

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Greece is phytogeographically a borderline and a crossroads of Europe and Asia, characterized by high plant diversity (6695 taxa) and endemism (22%). The high taxon diversity of Greece is consistent with a 300-year history of botanical exploration; this is one of the reasons that the Botanical Museum of the University of Patras (Herbarium UPA) was founded in 1973. The UPA Herbarium is traditionally focused on native plant taxa collections aiming to investigate and document the wealth of the natural heritage of Greece; it also constitutes the basis for systematic, floristic and biogeographical studies of the Greek plants and for teaching purposes. The aim of the present study is to highlight the scientific and cultural value of the UPA Herbarium due to its valuable botanical and genetic legacy of the Greek Flora. The UPA Herbarium hosts more than 70.000 plant voucher specimens of vascular plant which date from 1816 until today and are part of collections of Greek and European researchers from a range

of geographical regions (mountains, islets-islands, wetlands, grasslands, woodlands etc.). Herbarium collections include plant specimens belonging to 176 families and 1021 genera and representing more than 88 and 84% of the total of Greek Flora's families and genera, respectively. About 98% of them are Angiosperms, 0.6% Gymnosperms and 1.4% Pteridophytes. Most of the plant specimens come from the phytogeographical regions of Sterea Ellas (23.7%) and Peloponnisos (20.6%). Ionian Islands (10.4%) and Kriti & Karpathos (8.8%) are the most represented insular phytogeographical regions. About 40.3% and 32.8% of the UPA plant specimens represent mountain and insular floristic elements of Greece, respectively. The UPA Herbarium collections have been organized in accordance with the herbaria norms worldwide. Specimens are constantly added to the UPA collections and a mid-term target for our research group is to complete the inventory and digitization process of the deposited plant specimens of the rich in endemics Greek flora in order to make them available for researchers through the net.

**KEYWORDS:** Greek flora, Herbarium UPA, inventory, plant collections, plant specimens

Poster presentation 09 08 11

### TAU HERBARIUM: DIGITISATION OF SPECIMENS FROM THE EARLY ERA OF THE BOTANICAL EXPLORATION OF GREECE

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The Herbarium of Aristotle University of Thessaloniki (TAU) is a relatively small herbarium, which focuses on the Greek Flora and is associated with the Laboratory of Systematic Botany and Phytogeography, School of Biology. It is constantly enriched with specimens deposited by the Laboratory's current and former research staff and postgraduate students. However, it holds several old specimens, which date back to the early era of the botanical exploration of Greece. This work deals with the specimens collected up to 1850. A total number of 186 specimens date to this year (the oldest from 1830). The main collectors were: a) Theodor von Heldreich (1822-1902), German, one of the most prominent botanists and explorers of the Greek flora, who discovered more than 700 new species (65 specimens), b) Joseph Sartori (1809-1885), German, pharmacist in the court of King Otto (44 specimens), c) Theodoros Orphanides (1817-1886), the first modern Greek plant

scientist (15 specimens). The collection comprises 12 type specimens. Moreover, it includes 23 Greek endemic and 23 range-restricted taxa. The specimens' sheets and labels are left intact. The procedure includes a pre-digitisation stage, i.e. update of the taxonomy and nomenclature and addition on the cover of the specimen of two labels, the TAU barcode and the updated information label. The digitisation stage comprises imaging (photograph with a DSLR camera), georeferencing by using approximate collection locality, and databasing of the label information. All the above data will be incorporated to the GBIF research infrastructure in order to be made available online to serve education and research.

**KEYWORDS:** TAU Herbarium, Historical collection, digitisation of specimens

Poster presentation 10 08 10

### NOVELTIES FROM THE CARL STUDNICZKA'S HERBARIUM

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We analysed some families (Cannabaceae, Phytolaccaceae and Urticaceae; Ord. Canabineen, Empetreen, Phytolaceen and Urticeen) from the Carl Studniczka's herbarium (herbarium code NHMS). Most of the analysed herbarium material (35 herbarium sheets, with 65 samples of herborized plants) in this part of the C. Studniczka's herbarium were collected in Europe (27 herbarium sheets). According to the labels, the majority of herborized material was collected in the area of Italy (7 herbarium sheets). Most common herbarium sheets (7) are those from the Flora Dalmatiens collection. Studniczka himself (15) collected the majority of herbarium sheets. The oldest herbarium sheets date from 1867, whereas the newest one date from 1902. The exact year of collection is missing from six herbarium labels.

**KEYWORDS:** historical plant collection, Natural History Museum in Split, NHMS herbaria collection, Studniczka's herbarium

Poster presentation 11 08 02

### PLANTS PRESENT IN ALEXANDRU BELDIE HERBARIUM COLLECTED FROM THE BALKAN AREA

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Plants gathered from the Balkan area occupy an important role in the Alexandru Beldie Herbarium from „Marin Drăcea” National Institute for Research and Development in Forestry (INCDS) from Bucharest. This fact is proven by their scientific and historical value, both through the high variety of species contained, as well as through its large area from which the plants were collected for almost 150 years, an area that covers the entire Balkan region. The first part of the present paper shortly characterizes the Alexandru Beldie Herbarium, mentioning the most important species that it harbours, as well as the main personalities that have contributed to its development. The paper's main objectives are to systematize the main species collected from the Balkan area and present in the Herbarium, to describe the most important amongst them, including the rare ones as well as the oldest plants present in collections. Other objectives are represented by creating a map of the areas from which plants were gathered and emphasizing the periods with a maximum development capacity for the Herbarium collections. The assignation of objectives is then followed by the presentation of the material used, namely 318 plates that contain 189 plant species collected from the Balkan area. The work method is then delineated, focusing on the systematization of plates corresponding to the Balkan area, based on the species, gathering year and place and the person that has harvested each sample. Furthermore, an excerpt regarding the inventory of plants collected from the Balkans is also rendered. The paper's results and conclusions reunite, according to the established objectives, the description of the main Balkan species present in the Herbarium, as well as the results of systematizing the Herbarium's plates. As such, the periods of maximum development are rendered in a graphic method and described, followed by a map of all the Balkan areas from which the Herbarium's plants were collected. The personalities that have created this Balkan class of the Herbarium over 150 years are also mentioned, taking into consideration their professionalism in covering these extraordinary beautiful European areas in order to leave us an inestimable scientific and historical heritage.

**KEYWORDS:** herbarium, species, flowers, leaves, calice, botanists

Poster presentation 12 08 03

### QUERCUS SPECIES PRESENT IN THE ALEXANDRU BELDIE HERBARIUM

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*Quercus* is a reference genus of Al. Beldie's Herbarium from „Marin Drăcea” National Institute for Research and Development in Forestry (INCDS), Bucharest. This assertion is proven by the fact that it is one of the best represented species from the herbarium, as well as through its species diversity, harvest locations and the specialists that have gathered them over a long period of time. The present paper's introduction part overviews the main characteristics of Alexandru Beldie's Herbarium, presenting the main species that it contains in its collections as well as the personalities that have contributed to its creation. The paper continues with presenting the established objectives, namely systematizing the herbarium maps that contain *Quercus* species with the purpose of identifying the periods with a maximum development capacity and creating maps rendering the plant's harvesting areas. Another important objective is to identify and describe the most important *Quercus* species present in the Herbarium, as well as the rare, endangered or oldest ones. The work material is then presented, namely 1219 plates that contain *Quercus* species, as well as the work method used in order to describe the studied plates and systematize them based on species, gathering year and place and the person that has collected them. Together with the material and work methods, an excerpt of the Herbarium's *Quercus* genus inventory is also rendered. The results and conclusions reunite a graphical presentation of *Quercus* harvesting periods with a presentation of the harvesting location map. Furthermore, the most remarkable species of the Herbarium are also presented and described, together with the most representative, rare and old ones (dating back to 1850). The final part of the paper offers relevant conclusions regarding the most important aspects of the Herbarium's *Quercus* species, emphasizing their most remarkable features.

**KEYWORDS:** trees, collections, botanists, herbarium

Poster presentation 13 08 13

### VIHOTSEVSKI'S COLLECTION ON THE FLORA OF VITOSHA MT. (BULGARIA) KEPT IN THE HERBARIUM SOM

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The herbarium of the Department of Plant and Fungal Diversity and Resources of Institute of Biodiversity and Ecosystem Research, Bulgarian Academy of Sciences (SOM) is the largest and most representative source of specimen-based information about the Bulgarian flora as well as one of the most significant information centers for the Balkan flora. At present the herbarium comprises some 175000 specimens from all higher taxonomic groups of the embryophytes – bryophytes, lycopodiophytes, equisetophytes, polypodiophytes, gymnosperms and angiosperms. Of special interest are the specialized collections, which are also stored here. One of them is the thematic collection of the researcher Nikolay Vihotsevski on the plant diversity of the Vitosha Mt., near Sofia city, Bulgaria. Its thematic plant collection, stored in SOM and dedicated to the higher flora of the Vitosha Mt., consists of 421 herbarium specimens from 185 genera and 49 families, of which 1 equisetophyte, 12 polypodiophytes, 12 gymnosperms and 396 angiosperms (105 monocotyledons and 291 dicotyledons). The richest of genera are the following families: Asteraceae (87), Cyperaceae (24), Poaceae (75), Srophulariaceae (37), Lamiaceae (23), whereas the richest of species are the following genera: *Carex* (17), *Centaurea* (12), *Galium* (11), *Festuca* (11), *Veronica* (10), *Myosotis* (9), *Campanula* (9), etc. The collection includes some species with conservation concern: 2 species are Bulgarian endemics, 10 – Balkan endemics, and 3 are included in the Bulgarian Biodiversity Act. In conclusion, it is worth noting that the significance of the mentioned collection for the study of plant diversity of Vitosha Mt. and Bulgaria continues to be of immense importance also nowadays.

**KEYWORDS:** Bulgaria, Herbarium SOM, Vihotsevski, Vitosha Mt.

Poster presentation 14 08 08

### DIGITISATION OF DIMITRIOS ZAGANIARIS' HISTORICAL BOTANICAL COLLECTIONS DEPOSITED IN TAU (GREECE)

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Herbaria are deposits of biodiversity. Old botanical collections can be of equal importance compared to modern collections, since they provide information about past environmental facts, which may help scientists to assess the environmental changes in 21<sup>st</sup> century. The Thessaloniki Aristotle University Herbarium (TAU) holds several botanical collections from Greek and other European researchers dating back to the early 19<sup>th</sup> century. Dimitrios Zaganiaris (1900-1940) was a Greek botanist who travelled around Greece and published about eight floristic surveys between 1932 and 1940. One of his well-known published work is “Herbarium Macedonicum” (1938-1940), consisting of four volumes, with a total number of 4000 plant recordings from different regions of Northern Greece. However, many of his data were not made available to the scientific community, as they remained unpublished because of his early death, while several of his specimens were destroyed during World War II. This project aims to record all Zaganiaris' specimens found in TAU, targeting, through their digitisation, to make them accessible worldwide and useful to contemporary taxonomic and floristic studies. A pre-digitisation process has already been started, attempting, at first step, to verify which collections are published in any of Zaganiaris' floristic studies. Our preliminary results have shown that 2035 specimens are deposited in TAU, representing plant species of 112 families. Among them, 457 specimens correspond to records published in “Herbarium Macedonicum”. The ongoing digitisation process comprises: a) checking and updating nomenclature of all specimens, where necessary, b) extracting and abstracting label information, c) recording the collection locality of the specimens; the coordinates of the different regions will be defined by a graphical display software (e.g. “Google earth”), while older toponyms, some of which are Serbian or Turkish, will be matched to current toponymy, d) creating digital high-resolution images of all Zaganiaris' specimens. All the above information will be incorporated to a large biodiversity database, such as “Global Biodiversity Information Facility” (GBIF).

**KEYWORDS:** Zaganiaris, TAU Herbarium, Greece, historical collections

## Session 9. Ethnobotany



Balkan  
Botanical  
Congress

Introductory lecture 01 09 01

### MEDICINAL PLANTS OF THE BIBLE - PAST, PRESENT AND FUTURE

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Based on data on archaeobotany and ethnobotany of the Holy Land, survey of the use of medicinal plants in the Holy Land throughout history as well as at the present time, and a revision of the medicinal plant of Assyria we suggest a new list of the Medicinal Plants of the Bible. Only five species are mentioned directly as medicinal plants in the Bible: Fig (*Ficus carica*), Nard (*Nardostachys jatamansi*), Hyssop (*Majorana syriaca*), Balm of Gilead (*Commiphora gileadensis*) and Mandrake (*Mandragora officinarum*= *M. autumnalis*). While Duke and Duke (1983) enumerated not less than 176 plant species as "Biblical Medicinal Plants" and Jacob (1993) only 54, in our survey we suggest reducing that figure to 37. Not less than 18 medicinal plants, additional to the Bible, are mentioned in old Jewish post-Biblical sources. Most of these plants (15) are known also in Egypt and Mesopotamia while 3 only from Egypt. Seven of the BMP's species are not mentioned in the Bible or in the old Jewish Post-Biblical literature but were recorded as medicinal plants from Egypt as well as from Mesopotamia, and it is quite logical to assume that they can be included as BMP's. According to our survey, all the 37 suggested BMP's are still in medical use today in the Middle East and are subjected, at the 21 century, to an active research in attempts; to understand their chemical composition and/or medical activity and/or isolation of new compounds for new drug development. Shakya (2016) mentioned "Top 25 Bioactive Compounds of Medicinal plants", his list includes also: *Curcuma longa*, *Ricinus communis*, *Piper nigrum*, *Aloe vera*, *Nigella sativa*, *Artemisia absinthium* and *Allium sativa*=19% of our list of Biblical Medicinal Plants!! As written in the Bible: "That which has been is what will be, That which is done is what will be done, And there is nothing new under the sun". (Ecclesiastes 1:9).

**KEYWORDS:** Bible, medicinal plants

Oral presentation 02 09 10

### TRADITIONAL WILD FOOD PLANTS USED IN KNIN AREA, CROATIA

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The documentation of the traditional knowledge of wild edible plants use in Dalmatian hinterland is of great importance because the human population of that region is rapidly decreasing. The aim of this preliminary study was to record the use of wild plants as food by indigenous people in Knin area in south-eastern Croatia. Forty eight native inhabitants (38 female and 10 male) were interviewed during 40 semi-structured interviews. Presented data was extracted from broader research on plant use in the area. The uses and preparations of 85 plant taxa belonging to 37 families and 194 vernacular names from 630 citations were recorded. Lamiaceae, Compositae and Rosaceae were the most represented families, and the most commonly used wild plants for food were *Urtica dioica* L., *Thymus longicaulis* C. Presl, *Sambucus nigra* L., *Cornus mas* L. and *Rosa canina* L. Plants are predominantly used as vegetables and as infusion followed by syrups. The holders of the knowledge are older native inhabitants (median age was 71), while their decedents mostly moved out of the area. There is serious danger of loss of the traditional knowledge of wild plants use for food.

**KEYWORDS:** ethnobotany, wild edible plants, foraging, infusion, syrups

Oral presentation 03 09 02

### ETHNOBOTANY OF ARUMS IN TURKEY

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*Arum* taxa are seasonally dormant herbs with rhizomatous tubers. Leaves are sagittate or hastate. Male, female and sterile flowers are small usually crowded at the base of a spadix and generally encircled by a spathe. The fruits are bright glossy with orange red berries on a spike; the pericarp is juicy; seeds few to many per berry. Due to the wide range of variation and existence of many transitional forms, systematic positions of Turkish aroid taxa have been differently interpreted by vari-

ous authors. In accordance with various resources including relevant volume of the Turkish Flora it is reported by different authors that 14 or 17 taxa exist in Turkey. The conflict between the numbers of the existing taxa is due to the phenotypic variation. *Arum* species have been familiar to East Mediterranean people since antiquity and were used mainly as medicine and food. These taxa are still used by the traditional healers for the same purposes. However, due to significant amount of calcium oxalate crystals, oxalic acid, and oxalates as well as volatile and/or unstable irritating components *Arum* is considered as poisonous. The objective of the study is to discuss information on utilization of these plants from the ethnobotanical point of view. Major part of the ethnobotanical information comprising the vernacular names and way of uses was collected from the local people who know about these plants. The evidence for all kinds of uses for *Arum* taxa in Turkey from antiquity to current traditional medicine has also been summarized. The assessment of the compiled information directs the researchers to make more detailed studies on this complex group of plants including endemic species thereof so as to reveal the ethnobotanical value of Arums in Turkey.

**KEYWORDS:** *Arum*, Turkey, ethnobotany

Oral presentation 04 09 07

### DECORATIVE INDIGENOUS PLANTS ON ZAGREB FARMERS' MARKETS (CENTRAL CROATIA)

Vedran Šegota<sup>1</sup>, Nina Vuković<sup>1\*</sup>, Anja Rimac<sup>1</sup>, Tihana Vilović<sup>1</sup>, Nikola Koletić<sup>1</sup>, Ivan Limić<sup>2</sup> & Antun Alegro<sup>1</sup>

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The history of Zagreb farmers' markets began even before the establishment of the City itself, as early as 13<sup>th</sup> century, when the first public market was established in the area. The beginning of the today's market system was in 1930, when the central market Dolac was opened for public. After that time and especially after the World War II, the market system enlarged and spread following the spread of the City, and nowadays includes 22 locations. Apart from food and local domestic products, the sellers sometimes offer decorative flowers, out of which some are not cultivated but collected in natural habitats. Our aim was to prepare the overview of the decorative indigenous plants sold on Zagreb farmers' markets. We have studied all 22 locations in the period January-April 2018, by visiting the markets and surveying the sellers. We made several visits to the markets offering flowers, to include the whole spring season, while the data were collected using previously prepared, original questionnaires. Decorative indigenous plants were found on 13 locations. Total number of respondents was 50, mostly women of the average age 53.

Total number of recorded plant taxa was 79, out of which 39 herbs, 15 trees, 14 shrubs, eight mosses and three vines. The respondents provided 64 different common names for a total of 45 plant taxa, mostly giving one name per species, but sometimes also several names per species or the same name for several species. Altogether 93 % of plants were sold as cuttings, while 7 % plants were provided in whole. Only six plant species were provided with underground parts, for further planting. We found two legally strictly protected taxa, *Taxus baccata* L. and *Helleborus niger* L. subsp. *macranthus* (Freyn) Schiffner, both listed in the Red Book as vulnerable (VU). In general, our conversations with the respondents revealed that they are rather familiar and in agreement with the legal protection of the spring flora.

**KEYWORDS:** ethnobotany, spring flowers, local markets, ornamental plants

Oral presentation 05 09 15

### BARTIN'S NATIVE GEOPHYTES AND THEIR ECONOMIC IMPORTANCE

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The native geophyte taxa of Bartın, one of Turkey's rich flora areas were identified in this study. Bartın province located in the Western Black Sea Region of Turkey is between the eastern longitudes of 32<sup>o</sup>- 33<sup>o</sup> and the northern latitudes of 41<sup>o</sup>-42<sup>o</sup>. It is surrounded by Kastamonu in the east, Zonguldak in the west, Karabük in the southeast and Black Sea in the north. Altitude varies from sea-level to 1619 m. asl. in Bartın. According to Davis squaring system, the research area is within A4 square in the map of Turkey. It is observed mainly Euro-Siberian and partially Mediterranean flora characteristics in the research area. However, there are also some species of Iran-Turan flora region in the south of the area. Variability in topographic structure and soil properties, Oceanic bioclimate, the existence of sand dunes in sea coast, the presence of karstic fields in Kure Mountain National Park, deep valleys between mountain ranges, and riparian lands in Bartın have all a positive impact on its plant biodiversity. Site studies were carried out to collect plant samples in vegetation periods between 2011 and 2017. As a result, based on the taxonomic identification of 1800 plant samples, we determined 109 geophyte taxa belonging to the 57 genera and 26 different families, and 7 of them are endemic for Turkey. Many geophyte taxa in Bartın have been collected illegally from natural habitats because of their valuable bulbs, tubers, corms or rhizomes. The most common taxa illegally collected and market-

ed for economic gain are *Gagea bohemica* (Zauschn.) Schult & Schult., *Pancreatium maritimum* L., *Crocus ancyrensis* (Herb.) Maw., *Ornithogalum fimbriatum* Willd., *Anacamptis pyramidalis* (L.) Rich., *Orchis mascula* (L.) L., *Equisetum arvense* L., *Leucojum aestivum* L., *Lilium martagon* L., *Ruscus aculeatus* L., *Ruscus hypoglossum* L., *Asparagus aphyllus* subsp. *orientalis* (Baker) P.H.Davis, *Iris suaveolens* Boiss. & Reut. Uncontrolled and intensive plant gathering, uncontrolled grazing, intensive forestry applications and road network studies negatively affect the species diversity of geophytes. In-situ and ex-situ conservation measures should be taken especially for endemic and narrow-spread geophyte taxa under threat.

**KEYWORDS:** geophyte, flora, bulb, tuber, corm, rhizome

Poster presentation 06 09 13

### “VELIKI SRPSKI TRAVNIK “ - THE NEGLECTED HERITAGE

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In this paper we present and discuss the origin and scientific significance of two works that are kept in the collection “Zaharija Orfelin's Library” in the Patriarchate Library of the Serbian Orthodox Church in Belgrade. These are the first volume of “Veliki srpski travnik” (Eng. “Great Serbian Herbal”) and a newly discovered unidentified work of similar content. The aim of this study is to clarify the authorship of these works and bring them to the attention of the Serbian scientific community. In order to state the authorships precisely, all available literature on the subject has been reviewed. A comparison *de visu* of the copies of selected herbals from the Patriarchate Library in Belgrade, the British Library and the Royal Botanical Gardens at Kew in London, and at the Bavarian State Library in Munich has been conducted. For many years Zaharija Stefanović Orfelin (1726-1785), a Serbian versatile creator, has been credited with the authorship of “Veliki srpski travnik”, although it had been established as far back as in 1921 by Mita Kostić that its authorship is manifold. “Veliki srpski travnik” is clearly one of the copies of “Herbarium Blackwellianum”, created first as a “Curious Herbal” in 1735 by Elizabeth Blackwell (1707-1758), a British herbalist, and later redrawn by Nikolaus Friedrich Eisenberger (1707-1771), a German painter. Zaharija Orfelin bought a copy of the herbal and recreated it as a 3-volume work. For the first volume he assigned Serbian names for the plants to each of the 490 illustrated plates, and described the attributes and usage for 7 plants. Identification of a newly discovered work from the Patriarchate Library in Belgrade has not yet been confirmed for certain, as an ink analysis is needed to determine the time range of creation.

However, the plates, the illustrations and the handwriting of Serbian plant names imply that most probably it is one of the two lost volumes of “Veliki srpski travnik”. Even if he is not the author of herbal's concept and illustrations, Orfelin's contribution in terms of botanical nomenclature was a significant milestone for the foundations of Serbian botany.

**KEYWORDS:** history of botany, herbal, Zaharija Orfelin, Elizabeth Blackwell

Poster presentation 07 09 12

### NOTES ON ARCHAEOBOTANY, ETYMOLOGY AND LEXICOLOGY OF THE GENUS CAMELINA CRANTZ

Ana Marjanović Jeromela<sup>1\*</sup>, Johann Vollmann<sup>2</sup>, Aleksandar Medović<sup>3</sup> & Aleksandar Mikić<sup>1</sup>

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The genus *Camelina* Crantz, colloquially referred to as *false flax*, belongs to the family *Brassicaceae* Burnett (syn. *Cruciferae* Juss.) and comprises several herbaceous species originating in Near East, Asia Minor and South Europe. *C. sativa* (L.) Crantz, usually referred to as *camelina*, *false flax* and *gold-of-pleasure*, is one of the most ancient oil crops. The archaeological evidence of its presence in diverse ancient cultures and local agricultures is rich and enables tracing its spread throughout various ages and across the Old World. It is most likely that *camelina* followed common flax (*Linum usitatissimum* L.) during its spatial and temporal distribution, possibly adapting its growing habit to that of the latter and surviving as its weed-like companion. This may be confirmed by the etymology of the Greek word denoting *camelina*, consisting of *χώρα* (*ground*) and *λίναριον* (*flax*), depicting a lower competing ability of *camelina* when growing together with the flax crop. The material testimony of *camelina* ranges from Neolithic (eight millennia ago) to Roman and Medieval times and from Karakorum in Mongolia, over Armenia and Baltic coastline to the Iberian Peninsula. The common names denoting *camelina* in numerous ethnolinguistic families distinct the dark yellow or rusty colour of its flowers and seeds. This may be seen in modern languages, such as the Indo-European, with the German *leindotter*, the Italian *dorella*, the Russian *ryzhik* or the Sorbian *žoltk*, and the Uralic languages, with the Finnish *ruistankio* and the Hungarian *sárgarepce*. Some others, such as the Celtic Welsh, with *cydlin*, consider *camelina* a plant that is literally *with flax*. A vast majority of the Slavic languages regard *camelina* as something similar to flax and thus have their common names derived from those designating flax, such as the the Polish *lnicznik/len*, the Serbian *lanik/lan* or the Slovak *laničník/lan*. The vernacular names for *camelina* may be associated with oil, as seen in the Swedish *ol-*

*jedâdra*, or with other crops grown for the same purpose, such as in French, with *sésame d'Allemagne*. Merging archaeobotany and linguistics may cast more light not only on the present, but also on the earliest past of crops, including camelina.

**ACKNOWLEDGEMENTS:** This work is the result of research under the project TR 31025, financed by the Ministry of Education, Science and Technological Development of the Republic of Serbia.

**KEYWORDS:** archaeobotany, *Camelina sativa*, crop history, false flax, linguistics, oil crops

Poster presentation 08 09 05

### ETHNOGRAPHIC HISTORICAL SOURCES AS A PRELUDE TO ETHNOBOTANICAL RESEARCH IN PODHALE REGION

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Podhale is a cultural region in southern Poland, in the northern foothills of Tatra Mountains. It is a region of ethnographic and geographical uniformity, which for decades has not undergone the process of urbanization, while maintaining numerous customs and characteristics of traditional culture. Ethnographic research in this region has been conducted since the beginning of the 19th century. The main topics that interested researchers of the 19th and 20th centuries were shepherd's customs, architecture, clothing and dialects. Unfortunately, ethnobotanical studies have not been conducted in Podhale until today. This paper is an ethnobotanical analysis of historical sources on the ways of using wild plants by the inhabitants of Podhale. Here, over 25 ethnographic publications and 71 ethnographic interviews from the collections of the Tatra Museum of Natural Sciences in Zakopane, as well as archival interviews from the Polish Ethnographic Atlas and old manuscripts / guides, so-called "jottings" (a kind of guidelines leading to treasures hidden in Tatra Mountains and magical rituals allowing to find them), have been used. In addition, letters to Józef Rostafiński (from 1883) have been analyzed. He conducted a nationwide ethnobotanical survey, as a response to which, J. Rostafiński received almost 860 letters from about 370 respondents, and only two of them came from the Podhale region. The result of this analysis is a list of 177 plant species belonging to 62 families, which were used in the kitchen (mainly as food in hunger times), folk medicine, local architecture and furniture, as well as in magical rituals and religious ceremonies. These analyzes of historical ethnographic sources show that Polish highlanders have been using many of the plants growing in their immediate surroundings. Therefore, they will serve as an introduction to further ethnobotanical research that will be carried out in the near future in Podhale. The final list of species will be the basis for com-

paring the knowledge about the use of wild plants by Polish highlanders in the past and nowadays.

**KEYWORDS:** Podhale, ethnobotany, Polish highlanders, use of wild plants, Tatra Mountains

Poster presentation 09 09 08

### PLANTS AND SENSE OF IDENTITY – CULTURAL ECOSYSTEM SERVICES IN GOTSE DELCHEV MUNICIPALITY, BULGARIA

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Plants and vegetation are in unbreakable relation with all human activities. Because of these links numerous species and habitats were adopted as cultural symbols by the local communities. This survey is an attempt to describe the variety of plants and plant communities which are part of the sense of geographical, ethnic and religious identity of the population of Breznitsa and Banichan - villages in Gotse Delchev municipality in South West Bulgaria. Ethnobotanical methods such as semistructured individual interviews and focus groups were used to gather the information from the members of the local communities. An interpretation of the data was made considering the concept of the cultural ecosystem services which shows the importance of the agroecosystems, forest and grass ecosystems and the lands with sparse vegetation on defining the self-perception of the members of the rural communities.

**KEYWORDS:** cultural ecosystem services, sense of identity, ethnobotany, Breznitsa, Banichan, Bulgaria

Poster presentation 10 09 04

### TRADITIONAL USE OF MEDICINAL AND EDIBLE PLANTS ON STARA PLANINA (SOUTHEASTERN SERBIA)

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This study provides significant ethnobotanical information on medicinal plant use in the Stara planina region (south-eastern Serbia). The research area is characterized by a high diversity of plant species, which have a wide range of medicinal and dietary uses among the local population. The aim of this study was to document all the traditional knowledge and analyze the medicinal plants used in

this area, as well as to identify plant species of importance for future pharmacological studies. Local knowledge was obtained through semi-structured and open interviews, in which 51 informants, aged between 49 and 92 (with a mean age of 70.5), were interviewed. The relative importance of the plant species was determined by calculating the use value (UV). The informants provided data on 157 medicinal and edible plants belonging to 57 families, of which *Asteraceae*, *Lamiaceae* and *Rosaceae* predominated in their local use. The species with the highest use values in ethnobotany and diet were *Allium ursinum*, *Achillea millefolium*, *Carlina acaulis*, *Cornus mas*, *Corylus avellana*, *Fragaria vesca*, *Gentiana asclepiadea*, *G. cruciata*, *G. lutea*, *Hypericum perforatum*, *Juglans regia*, *Mentha × piperita*, *Plantago lanceolata*, *P. major*, *Rosa canina*, *Rubus fruticosus*, *R. idaeus*, *Sambucus nigra*, *Satureja montana*, *Thymus serpyllum*, *Vaccinium myrtillus* and *V. vitis-idaea*. Medicinal plants were used most commonly to treat respiratory, gastrointestinal, urogenital, skin and cardiovascular conditions, as well as for detoxification and strengthening the body. Aerial parts of medicinal plants (mostly when in bloom) are traditionally used in making various preparations (teas, decoctions, tinctures, oils, ointments, balms, juices, syrups, and 'travarica' brandy). Ethnobotanical research in the Stara Planina region has established that the fruits, leaves, aerial parts or roots of the 47 plant species are used as food and drink in the form of juices, syrups, sweets, brandy, spices, salads (in their fresh state) and for making various other dishes. Ethnobotanical knowledge in this area is decreasing due to high emigration rates in recent times. However, this historically developed ethnobotanical heritage should be preserved and promoted on a wider level and given special consideration in future management plans for the Stara planina region.

**KEYWORDS:** ethnobotany, Stara planina (Serbia), medicinal plants, edible plants, traditional plant uses

Poster presentation 11 09 14

### ETHNOBOTANIC STUDY OF SOUTHERN ŠAJKAŠKA

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Sajkaska is a region in province of Vojvodina, northern Serbia. Region is located in humid continental climate zone. This is flat land transected with big rivers. Because of that, this region is very suitable for agriculture. This paper presents a research in southern part of Sajkaska, region located in Novi Sad and Titel municipalities and 9 settlements: Kac, Budisava, Kovilj, Sajkas, Mosorin, Vilovo, Lok, Gardinovci and Titel. The aim of this paper is to assume diversity of economically important flora of Southern Sajkaska region. This analysis is

conducted by collecting data from local population. In every settlement 30 people was polled and they were answering questions about fruit, vegetable, grain, spice and medicinal plant species which they grow, as well as plant species that were grown in the past and also those that are planned to be grown in the future. Collected data were analyzed with classical statistical methods. Results prove correlation between number of different plant species and microclimatic characteristics of settlements, cultural differences between ethnicity of polled people, education levels, type of jobs of people and their age. There are certain trends in types of plants that are grown in different settlements. Also, there are strong correlation between diversity of grown species and resident's age. Considering education levels there are trends indicating higher diversity of grown vegetable, spice and medicinal plant species and species planned for growing in the future. Analysis of resident's ethnicity, indicate higher plant diversity in settlements with more different nationalities living together. This research presents trends in selecting and growing different plant species. It shows trends and correlations about geography and ecology of region and their influence in forming specific groups of species region. Researches like this have great importance in estimation of economically important flora of the settlements and also ethnological and ethno-botanical importance. Also, they compile informations about rare and forgotten species and cultures of researched area.

**KEYWORDS:** cultivated plants, economical importance of plants, Šajkaška

Poster presentation 12 09 03

### USING ELLENBERG-PIGNATTI VALUES TO ESTIMATE HABITAT PREFERENCES OF WILD FOOD AND MEDICINAL PLANTS: AN EXAMPLE FROM NORTHEASTERN ISTRIA (CROATIA)

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The paper presents the first ethnobotanical application of Ellenberg indicator values, which are widely used in European plant ecology. The aim of the study was to find out if Ellenberg values (indicating habitat preferences) differ for wild food and medicinal plants used in north-eastern Istria (Croatia). We used Ellenberg-Pignatti values (the version of Ellenberg values used in this part of Europe). Fifty semi-structured interviews were carried out among local key informants, asking which wild food and medicinal plants they used. The mean number of food and medicinal plants mentioned per interview was 30. Altogether, 121 species were recorded as food

or medicine used or previously used in the study area. Thirty-one species are used exclusively as food or everyday drink, 50 species are used exclusively as medicine and 40 species are used for both food and medicine. The most commonly used exclusively food species are: *Cornus mas*, *Cichorium intybus*, *Chenopodium album*, *Prunus domestica*, *Pyrus amygdaliformis*, *Rubus idaeus*, *Clematis vitalba*, *Diplotaxis tenuifolia*, *Fragaria vesca* and *Allium ampeloprasum*. The most commonly exclusively medicinal species are: *Achillea millefolium*, *Tilia platyphyllos*, *Hypericum perforatum*, *Sempervivum tectorum*, *Artemisia absinthium*, *Plantago lanceolata*, *Gentiana lutea* ssp. *symphyandra*, *Althaea officinalis*, *Matricaria chamomilla*, and *Pinus nigra*. The most commonly used food-medicine spectrum species are: *Rubus caesius*, *Sambucus nigra*, *Urtica dioica*, *Dioscorea communis*, *Taraxacum* spp., *Asparagus acutifolius*, *Rosa canina*, *Foeniculum vulgare*, *Prunus spinosa* and *Sorbus domestica*. There were no significant differences between Ellenberg values for food and medicinal plants, apart from the Nitrogen indicator value – the plants used exclusively as food had a significantly higher index than those used in medicine. This probably stems from the fact that plants with soft fleshy shoots are attractive as food and they are more likely to come from nitrogen-rich ruderal habitats. Food plants and medicinal plants are collected from a variety of habitats and no clear difference between the two categories of plants was detected, however further testing of Ellenberg values in ethnobotanical studies could be interesting.

**KEYWORDS:** ethnoecology, quantitative ethnobotany, Ellenberg indicator values, wild edible plants, medicinal plants, Croatia

Poster presentation 13 09 11

### HYPERICUM PLANTS AS TRADITIONAL MEDICINES IN AREAS OF NORTH CENTRAL AND EAST PELOPONNESE (GREECE)

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The genus *Hypericum* L. includes about 484 taxa (herbs, shrubs and infrequently trees) placed in 36 taxonomic sections with worldwide distribution in warm temperate, subtropical and mountainous tropical regions. Several publications suggest that wild growing *Hypericum* plants are well known in the Balkans and are used by local people for the preparation of traditional medicines. The genus is represented in the flora of Greece by over 40 taxa (species and subspecies), members of 13 sections. Among them 15 taxa are Greek endemics. *Hypericum* plants are widely collected from the Greek country under the name “spathohorto” and “valsamohorto”

and are used for the preparation of teas or more frequent of olive oil extracts (“spatholado” or “balsamolado”). The aim of the present study is to (i) collect and identify the different *Hypericum* taxa grown in two geographically distant regions of Greece, (ii) record which of them are used as traditional medicines by the locals, and (iii) trace information for the way of home-made preparations as well as the suggestions for therapeutically applications. The total area where the present study is carried out, includes 12 villages and small cities, six located in North Central Greece (Municipality of Edessa) and six in the Eastern part of Peloponnese (Municipality of Epidaurus). Plants of the genus *Hypericum* were collected during flowering time and taxonomically identified. Voucher specimens with coordinates of their collection sites, are kept at the Herbarium of Aristotle University of Thessaloniki (TAU). For the collection and evaluation of ethnobotanical information, structured and semi-structured interviews with native people were used. The informants are of different sex, age and professional employment. Due to differences in topography and climate (Continental-Mediterranean and Mediterranean climatic zone, respectively) but also in the cultural heritage of the two areas, differences are recorded in the taxa used as well as in the way of their therapeutically uses.

**KEYWORDS:** *Hypericum*, ethnobotany, taxonomy, traditional medicine, Greece

Poster presentation 14 09 06

### DIVERSITY OF RARE INDIGENOUS PEAR VARIETIES (*PYRUS COMMUNIS* L.) IN THE REGION POLIMLJE, (SOUTHWEST SERBIA) AND THEIR USE IN ETHNOMEDICINE AND NUTRITION

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This study documents the ethnobotanical and ethnomedicinal knowledge of the local inhabitants in the region Polimlje in Southwest Serbia, focused on recording rare autochthonous pear varieties (*Pyrus communis*). A special feature of Polimlje, as an unique multiethnic complex of natural and cultural heritage, is the great diversity of indigenous varieties of pears and breeding tradition in the households for hundreds of years. Objectives of the study were: to establish the diversity of indigenous pear varieties in order to preserve the unique *P. communis* gene fond characteristic to this region, and to explore their use in ethnomedicine and traditional

food production. The opened and semi-structured interviews were done with 40 people aged between 30-80 years (29 men and 11 women), during June - September 2015, on the territory of 23 villages. The results of this study indicate the presence of 23 autochthonous pear varieties: 7 are determined as rare (*Vidovača*, *Ječmenjača*, *Ilinjača*, *Lubeničarka*, *Medunak*, *Zimnjača*, *Takiša*) and 16 as extremely rare (*Jagodarka*, *Mirisavka*, *Sijerak*, *Turundžija*, *Stambolka*, *Čadavica*, *Okruglica*, *Mesnjača*, *Jarac*, *Bazva*, *Tepavac*, *Lončara*, *Kantaruša*, *Ovčara*, *Turšijara*, *Budaljača*). A lot of varieties remain in the old and abandoned homes and the rugged terrain. For medicinal purpose pear is used (fresh or dried fruit, juice or compote) as antihypertensive remedy (65.2%), antidiabetic or anticholesterol (63%), anticonstipation remedy (54.3%), as well as for body mass reduction (65.2%). Tea made from the pear skin is used for urolithiasis - stone and sand in the kidney (36.9%), pear tea and fresh fruit juice are applied as antirheumatics (26%) and for cancer prevention (10.87%). Pears are consumed as fresh fruit, in pies and cakes, or they are processed in a compote, juice, jam or marmalade, baby food, jelly, syrup, dried fruit, pear brandy and other traditional products (*sita*, *vodnjika*). Depopulation of investigated rural area is directly related to disappearance of pear varieties gene pool, thereby losing traditional knowledge. Preservation can be achieved with *in situ* methods, by raising the awareness of the inhabitants and through the support of the State.

**KEYWORDS:** *Pyrus communis* L., indigenous pear varieties, ethnomedicine, nutrition, region Polimlje (Southwest Serbia)

Poster presentation 15 09 09

### HOME GARDENS – GEOGRAPHY OF BIOCLTURE AND QUALITY

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The current study focuses on plant diversity used in production of traditional food from the Balkans. The selected products are part of Ark of Taste e-catalogue of Slow Food and their recognition and promotion is a result of the collaborative network of 8 Balkan countries (including Turkey). All entries involve small-scale farmers and processors engaged in preservation of food biodiversity and traditions through sustainable use of biological resources. From over 250 products (primary and processed), 174 were found to include plant ingredients or to be local varieties/landraces. Most of the latter are cultivated in gardens and/or as small-scale crops being part of the disappearing traditional practices handed down from generation to generation. About half of the products are manufactured by small businesses that offer food at local or regional markets and/or restaurants. Analyzing the threats for food diversity in the Balkans we have detected a high level of similarity that presumes common approaches to safeguarding it.

To illustrate the overall conclusions we show-case Bulgarian traditional products with geographical reference. Challenges and transformations related to plant heritage conservation at home gardens are discussed.

**KEYWORDS:** rural areas, plant varieties, agrobiodiversity, local communities

Poster presentation 16 09 16

### RISING INTEREST IN MEDICINAL PLANTS APPLICATION – DATA FROM NOVI SAD AND SURROUNDING AREAS

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The trend of self-medication and intense public promotions of herbal medicines and herbal dietary supplements sometimes results in use of plants of distant cultures, as well as use in unorthodox types of application. Phytopreparations with herbs previously not familiar to wider consumers market seem to be used together with plant of traditional herbal medicine. Purpose of the study was to determine the most frequently used medicinal plants in the territory of city of Novi Sad and suburban and rural areas surrounding the city. A questionnaire was specifically created for this research. Survey contained questions about sociodemographic characteristics and use of phytotherapy. The semi-open list with 64 medicinally significant plants was provided; the reasons of plant's usage were added in separated open list. Volunteers (age over 18 years) filled in the survey. The data were analyzed using IBM® SPSS® Statistics 20. The research retrieved 104 completed surveys. The most frequently used plants belong to traditional Balkan and European phytotherapy were chamomile (47), garlic and parsley (44), mint (42), propolis (31), sage (30), nettle (29), oregano and rose hip (25), basil and St. John wort (22), marshmallow (21), yarrow, flax and lemon balm (17), lime tree (16), caraway, winter savory and elder tree (15), comfrey, pot marigold and rosemary (14). However, some other species, originating from other continents, such as green tea (34), ginger (31), chokeberry and bearberry (30), aloe vera and cranberry (28), as well as cinnamon (22) are also frequently used. The main reasons of use were: improvement of immune system (28), problems of urinary tract (27), diseases of mouth and gums and common cold (16), as well as sedatives, hypnotics (11) and for gastrointestinal ailments (10). Medicinal plants, especially aromatic species, appear to be used most frequently in studied area. Many of the consumed plants are traditionally accepted and used for longer period of time, although some new species seem to find their position in prevention and treatment of various diseases.

**KEYWORDS:** herbal medicine, medicinal plants, phytotherapy

## Session 10. Cryptogam Biology



Balkan  
Botanical  
Congress

Introductory lecture 01 10 10

### BRYOPHYTE SANCTUARY (BBGB COLLECTION): NEEDS, PROBLEMS, TRENDS AND ACHIEVEMENTS

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Bryophytes are recognized in many conservation initiatives, having in mind that it is the second biggest group of terrestrial plants (after angiosperms). Worldwide, there are different achievements in conservation of protection of bryophytes. However, these are often parts of wider actions and often it terminates with the publication of red-lists or red data books. Although, there was examples of active conservation, these are rather exception. Saving the target species cannot be achieved by legislative only and some active help and measures are needed in many cases. Beside, in Global Strategy for Plant Conservation, Agenda 2010, or CBD, *ex situ* conservation and active protection is regarded also for bryophytes. However, the goal of at least 75 per cent of threatened plant species in *ex situ* collections, preferably in the country of origin, and at least 20 per cent available for recovery and restoration programs is not achieved yet. Also, this is not the case even in developed countries where the pressure to nature is rather high even though most of the world states ratified the document. The world biggest *in vitro* (i.e. *ex situ*) bryophyte collection with over 260 species from all over the world is harbored in the Botanical Garden of the Belgrade University and (Bryophyte Biology Group Belgrade - BBGB collection) and rather many activities are ongoing in active protection of bryophytes including conservation ecology, conservation genetics and emerging field of conservation physiology. The collection counts some 59% of species of high regional or global conservation interests. Laboratory researches, propagation, acclimation with goals like native population strengthening or reintroduction to the region of extinction are often supported by field studies with aim to save target species, population(s) or genetic entities of bryophytes. Needs, problems, trends and achievements will be presented and discussed.

**KEYWORDS:** mosses, liverworts, protection, collection, *ex situ*, *in vitro*

Introductory lecture 02 10 15

### EXPLORATION OF THE BALKAN BRYOPHYTE FLORA WITH A SPECIAL ATTENTION TO THE RARE AND THREATENED SPECIES

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In the last 15 years the Bryological Collection of the Hungarian Natural History Museum (HNHM) has dealt intensively with the Balkan area, from where there is still relatively few bryophyte data compared to other parts of Europe. Several joint field trips with SE European bryologists were organised to fill the gap. Special attention was paid to the habitats of rare, threatened species by visiting suitable sites, making collections, listing the bryophyte flora, drawing up the distribution and estimating population size of rare species. More than 17.000 specimens have been deposited in the HNHM collected during these field trips and 75 papers have been published on the basis of the material. Voucher specimens of 603 species can also be found in HNHM, which were reported for the first time from the various countries of Balkan. From conservation aspects wetlands are the most important habitat types in SE Europe, threatened mainly due to climate warming. Their extension is decreasing, and they are continuously losing the sensitive bryophytes of their species pool. Although the suitable bogs are small and sporadic in the Balkans, they still maintain several populations of wetland species of European conservation interest, such as *Campylium elodes*, *Drepanocladus polygamus*, *Hamatocaulis vernicosus*, *Scorpidium scorpioides*, *Tomentypnum nitens*. The alpine grasslands are also threatened due to climate warming. The Balkan high mountain areas still preserve very diverse bryoflora. Especially areas with acidic bedrock in high elevations have special conservation interest, because they are rare and scattered in the predominantly calcareous Balkan mountains. The bryophyte flora of acidic soil and siliceous outcrops contains many calcifuge species regarded as rare in this region and redlisted in many SE European countries, e.g. arctic, alpine leafy liverworts such as *Lophozia wenzelii*, *Marsupella sphacelata*, *Scapania praetervisa*, *Solenostoma confertissima* or saxicolous species like the members of Grimmiaceae family (*Grimmia caespiticia*, *G. reflexidens*, *Schistidium papillosum*). In shaded limestone rocky habitats the bryophyte assemblages also contain several species of European conservation interest, e.g. *Anomodon rostratus*, *Mannia triandra*, *Myurella sibirica*, *Taxiphyllum densifolium*.

**KEYWORDS:** liverworts, mosses, species of European conservation interest

Oral presentation 03 10 28

### NEW CHECKLIST OF BRYOPHYTES OF CROATIA WITHIN FLORA CROATICA DATABASE

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The most recent check-lists of Croatian bryoflora were published more than a decade ago (15 years ago for hepaticae, and 12 years ago for mosses). More recently however, major progress was made in the research of Croatian bryoflora, with many significant contributions in the field up to date. Therefore, the need to critically revalue the existing lists, and prepare a new list to be incorporated into the Flora Croatica Database (FCD) occurred in recent times. For this purpose, we analysed all available literature about the Croatian bryoflora, with a special emphasis on old, 19<sup>th</sup> century literature that was partly omitted from the existing check-lists. Additionally, we have analysed the collection of bryophytes from the Hungarian Natural History Museum. As a result, we have added approximately 25000 literature and 6000 herbarium records into the FCD, creating a list of altogether 782 taxa. Finally, the FCD continues to be regularly updated with additional data from ZA and ZAHO herbarium collections, which includes adding the photographs and synonyms of corresponding taxa.

**KEYWORDS:** Croatia, bryophytes, mosses, liverworts, checklist

Oral presentation 04 10 01

### PRELIMINARY RESULTS OF STUDY ON BRYOPHYTE ASSEMBLAGES FROM SELECTED SPOIL HEAPS IN SLOVAKIA

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Spoil heaps, abandoned heavy metal loaded sites, host interesting flora decades or centuries after their creation. Previous attention was given to spontaneously grown vascular flora. Here, we made an attempt to document and study bryophyte assemblages at two such sites (Malachov – Velká studňa and Lubietová – Podlipa) in the vicinity of Banská Bystrica (Central Slovakia). In total, 120 bryophyte species were recorded, out of which 41 are common for both spoil heaps, and 44 and

35 unique for Malachov and Lubietová, respectively. Based on 102 relevés made in Malachov, 68 species were combined in various assemblages within 22 physiognomic vegetation types. Here, the most abundant were assemblages dominated by *Brachythecium rutabulum*, *B. salebrosum* and *Calliargonella cuspidata*, indicating eutrophic and rather unstable situation. Sporadically, other species (e.g. *Tortella inclinata*) that dominate the surface of the spoil heap area, imply unequally distributed resources available, which influences a rather less clear community grouping. At this spoil heap we also recorded *Drepanocladus polygamus* (EN – Endangered in Slovakia) in a mesophilous grassland dominated by *Carex flava*, and *Trichostomum crispulum* (VU – Vulnerable in Slovakia) in a mixed sparse shrubland with several pioneer trees and mostly *Calamagrostis epigejos* in the undergrowth. In Lubietová, we recorded 64 species combined in at least 6 assemblages (based on 79 relevés in 10 physiognomic vegetation types). An obvious assemblage is the one with *B. rivulare* (alliance *Brachythecion rivularis*), but also a wet stable assemblage with *Aulacomnium palustre* and a rather moderately dry assemblage with *Ceratodon purpureus*. The evidence collected at this site suggests that situation is less variable from the other studied spoil heap. Well established bryophyte communities can be seen, at least in some parts of the spoil heap area. Additionally, some rare and threatened species were recorded as well. *Helodium blandowii* (EN – Endangered in Slovakia) was present in wet and rather eutrophic parts according to the species growing in its vicinity. *Coscinodon cribrosus* (CR – Critically Endangered in Slovakia) was found in the tree stand with *Pinus sylvestris* on the steep spoil heap body with coarse-grained waste rock.

**ACKNOWLEDGEMENTS:** The study was supported by the Scientific Grant Agency of the Slovak Republic under Grant VEGA 2/0040/17.

**KEYWORDS:** bryophytes, assemblages, spoil heaps, Malachov, Lubietová, Slovakia

Oral presentation 05 10 04

### BRYOPHYTES OF SERBIA - PROGRESS, UPDATED CHECK-LIST AND ANNOTATIONS

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During modern period of bryological research in Serbia numerous floristic studies were carried out. Some of the localities were already explored historically, but many previously unexplored areas were also studied. This has brought valuable new data on distribution of the known species as well as many new country records. Bearing in mind this fast progress in exploration of Serbian bryoflora, all accessible data concerning their occurrence in Serbia were compiled into Bryo database. The nomenclature was unified and new updated check-list

for hornwort, liverwort and moss species was done. Newly formed Bryo database enabled critical approach of the taxa on the list. Additionally, revisions of targeted taxa were carried out. As a result, some of the species are rejected from the territory of Serbia, while others are marked as doubtful reports or taxonomically unclear taxa, hence more thorough studies in the future are needed. Although Serbia is considered to be rather rich in bryophyte diversity, it is shown that its territory and many taxa are still unevenly surveyed. Compiled data represent strong basis for further research that should be focused on field exploration of lesser explored areas and important and rare bryophyte species, as well as their assessment, conservation and monitoring.

**KEYWORDS:** bryophyte research, rejected taxa, doubtful taxa

Oral presentation 06 10 33

### BRYOPHYTES OF THE FRUŠKA GORA MOUNTAIN (SERBIA)

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Fruška gora is a mountain in the southern part of Pannonian plane. It is mainly covered with forest vegetation, and characterized by dense hydrological network, hence bryophytes are often conspicuous elements of different vegetation types. According to the literature data and field researches during 2013-2017, total number of 193 moss, and 25 liverwort taxa was recorded. Liverworts belong to 16 families and 17 genera and represent about 18% of Serbian liverwort flora, while mosses belong to 27 families and 94 genera, and represent about 30% of Serbian moss flora. The families with highest number of taxa were: Brachytheciaceae, Pottiaceae, Hypnaceae, Amblystegiaceae, Mniaceae and Polytrichaceae, while the most abundant genera were: Brachythecium, Didymodon, Syntrichia, Hypnum and Orthotrichum. 21 taxa (16 mosses and 5 liverworts) is on the Bryophyte red list of Serbia and Montenegro (“critically endangered”-2; “endangered”-5; “vulnerable”-7; “low risk”-6; “data deficient”-1); three species are in the Red Data Book of European Bryophytes (ECCB), while 13 species are candidates for the New Red Data Book of European Bryophytes. Due to the facts that Fruška gora is region with high biodiversity, dense hydrological network, and a very complex geology, there are a lot of different microhabitats which are conducive to some rare bryophyte species. Ellenberg’s ecological indices for light, temperature, humidity and soil reaction were analyzed. The majority of species were acidophilous, and ones which prefer half shade, lower temperatures and dry to moderately humid habitats. Fruška gora is characterized by large habitat diversity (different types of deciduous forests, dry and wet meadows, loess cliffs, springs, water accumulations etc.). Despite the high anthropogenic in-

fluence on Fruška gora, forests and spring areas on this mountain are still characterized by high level of bryophyte diversity. Dominance of deciduous forests with variable proportion of different tree species (eg. *Fagus sylvatica*, *Tilia* spp., *Acer* spp. etc) is very important for diversity of epiphytic species. Also, several loess cliffs on Fruška gora slopes, are very important habitats for bryophytes, according to the fact that cryptogams are major plant component on these sites.

**KEYWORDS:** mosses, liverworts, Fruška gora, threatened bryophytes

Oral presentation 07 10 17

### REDISCOVERY OF LIVERWORT *MANNIA* *TRIANDRA* IN SLOVENIA

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*Mannia triandra* (Scop.) Grolle is a small thallose liverwort from the family Aytoniaceae. In Slovenia, the thriving of this protected species had been a mystery for more than a century, with no confirmations of its presence recently. In April 2017, the species was re-discovered in Soča valley in Southern part of Julian Alps. This finding raised questions about its actual distribution, particularly since the species is of European nature-conservation concern, listed in the Annex II of the Habitats Directive, and has its *locus classicus* in Slovenia (near Idrija). The aim of the present study-in-progress is to collect distributional data for *M. triandra* in the field and to evaluate the main characteristics of its habitats. The old data about the presence of the species in Slovenia are numerous, mostly in Alpine and pre-Alpine phytogeographical regions. In spring 2018, we systematically examined 5 previously known localities of the species and a number of potentially suitable habitats in order to gain a better insight into *M. triandra*’s distribution. So far, we confirmed *M. triandra* in 4 of the old localities examined, and found it in additional 3 new localities. These preliminary results show, that at least in some parts of the territory of Slovenia, the species is more common than it was assumed. The species was probably overlooked, since it’s a very small, short-lived and can only be identified in the first half of the spring. In most of the confirmed and newly discovered localities, *M. triandra* grows on moist sand in the shelter of (sun-exposed) dolomite or limestone rocks, which are overgrown with thermophilous vegetation, often together with another liverwort, *Preissia quadrata* (Scop.) Ness. The final goal of the present study is to prepare an expertise for including the species in the Natura 2000 protection areas in Slovenia.

**KEYWORDS:** *Mannia triandra*, liverwort, Slovenia, nature conservation, Natura 2000



Oral presentation 08 10 26

### DISCONTINUITY IN RESEARCH OF MACROALGAE IN CROATIAN FRESHWATERS – THE IMPLICATIONS OF THE GAP BETWEEN HISTORICAL AND RECENT KNOWLEDGE

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First “tips” about the macroalgal flora in Croatian freshwaters were provided during mid- and late-19<sup>th</sup> century, in the form of the observations of certain species of Chlorophyta, Rhodophyta and Chrysophyta. During the mid-20<sup>th</sup> century, there was a major breakthrough in this topic when a large number of papers dealing with the occurrence and abundance of algal flora in the Croatian watercourses was published. Great deal of these papers was published as offprints, in local language in local journals, many of them still available only in printed forms; while only a small number is scanned and available online. In addition, online scans often do not appear in traditional internet search engines, because they do not contain digital keywords recordings but are saved as textual photographs. Between these historical records and recent studies of freshwater algae, there was a large gap in surveys and publishing covering this topic in Croatia. Recent research dating from the last quarter of 20<sup>th</sup> century is mostly related to Characeae, as a result of particular professional interest and as a part of national biomonitoring scheme and specific freshwater macroalgal Chrysophyta, Phaeophyta and Rhodophyta species. Preliminary observation shows the tendency of recent papers to overlook some of the old information; not only by the authors but also by the paper reviewers. We conclude that the historical information was “lost” through time due to the hardly accessible literature (local papers printed in local language and in unspecialized journals) followed by a discontinuity in research. This overview provides a survey of macroalgal research through historical, hardly accessible literature, and links the results with the recent macroalgal research, therefore, we bring some recent information about occurrence of *Hydrurus foetidus* (Chrysophyta); *Heribaudiella fluivatilis* (Phaeophyta); *Bangia atropurpurea*, *Hildenbrandia rivularis*, *Lemanea* spp., and *Thorea hispida* (Rhodophyta) in Croatian freshwaters.

**KEYWORDS:** offprint, digital record, Rhodophyta, Phaeophyta, Chrysophyta, review

Oral presentation 09 10 18

### CALCIUM OXALATES IN LICHENS. THE POTENTIAL ROLE OF SQUAMULES IN DETOXIFICATION STRATEGIES

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Lichens represent a unique form of symbiosis between photosynthetic and fungal partners. They belong to one of the most sensitive organisms at the level of ecosystem pollution. Atmosphere deposition is the major source of mineral nutrition for lichens but in some specific situations, the substrate plays also an important role in the accumulation of mineral elements, including essential metals like calcium. Calcium is important for structures in cell walls as well as in biological membranes and it is an essential intracellular messenger in the cytosol. Oxalic acid is usually associated with calcium in lichens where it leads to the formation of calcium oxalate crystals. There are two forms of calcium oxalates in lichens: weddelite and whewellite. However, the localization of calcium and the identification of oxalate forms in lichens is largely unknown. In a survey of travertine localities (calcareous soils) in Slovakia, we found two lichen species *Cladonia foliacea* and *Cladonia furcata* (cladoniaceae). Energy-dispersive X-ray spectroscopy (EDX) revealed calcium uptake from the soil and its localization in the thalli of both species. Using a diffractometer and according to JCPDS-PDF2 database, and raman spectroscopy we can distinguish two calcium oxalate forms. Squamules are small leaf-like scales on some lichen species which serve as a taxonomic sign. The role of squamules is still not fully explored. Both species contained squamules, where the concentration of Ca was highest in thalli. This localization and accumulation of calcium oxalate in lichen thalli is an essential contribution to better understand calcium distribution and function in lichens.

**KEYWORDS:** lichens, calcium accumulation, detoxification, EDX, raman spectroscopy, X-ray

Oral presentation 10 10 30

### DISTRIBUTION OF INDIVIDUALS IN POPULATIONS OF BASIDIOMYCETES MYCETINIS ALLIACEUS, MARASMIUS ROTULA AND GYMNOPUS ANDROSACEUS FROM SERBIAN AND MONTENEGRO FORESTS

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Saprotrophic basidiomycetes play an important role in leaf litter decomposition, which is especially emphasized in nitrogen-limited boreal and temperate forests. Little is known about spatial distribution the size of individuals of this group of fungi. *Mycetinis alliaceus*, *Marasmius rotula* and *Gymnopus androsaceus* are widespread and usually produce numerous sporocarps, which makes them a good model for population studies of litter-exploiting basidiomycetes. Samples examined in this study have been collected from three localities in the forests of Republic of Serbia and Republic of Montenegro. Between 12 and 23 sporocarps from each analyzed species were mapped and collected from these sites. In order to determine genotype distribution at each of the three investigated sites, ISSR analysis was conducted using the primers (GTG)<sub>5</sub> and (GACA)<sub>4</sub> separately. On each site seven to fifteen genets were identified. The majority of detected genets were represented by a single sporocarp so it was not possible to estimate their size. The size of the genets with two or more sporocarps was estimated to be from 0.3 to 4.0m. Results presented in this study suggest that populations of these three species can consist of numerous and relatively small genets. Although relatively small samples of sporocarps were analyzed in this study, obtained results are important since they present some of the first data about population structure of litter exploiting basidiomycetes.

**KEYWORDS:** (GACA)<sub>4</sub>, (GTG)<sub>5</sub>, ISSR, litter-exploiting

Poster presentation 11 10 05

### PHYTOGEOGRAPHIC TRAITS OF SERBIAN BRYOFLORA

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Serbia has many gaps in bryological exploration of certain areas of the country and lacks extensive bryophyte collection.

New floristic studies during modern period of exploration and gathering all known data on distribution of bryophyte species in Serbia enabled geographic aspect analyses of bryophyte flora of Serbia for the first time. Relatively small surface of Serbia is characterized by several climate types as well as diverse relief, geology and vegetation, which explains its species richness and ecological divergence. It is shown that spectrum of areal types of bryophytes in Serbia is similar to that from the area of Southeastern Europe, although some distinctions are present. Dominant areal types are boreal (31% in Serbia and 24% in SE Europe) and oceanic (23% in Serbia and 25% in SE Europe), while least present is continental areal type (3% in Serbia and in SE Europe). Bryophyte flora within various areas of Serbia is in accordance with the environmental conditions (mostly macro climatic) and expresses different patterns and biogeographical peculiarities. For example, with the decrease in mean annual precipitation going from western to the eastern part of the country, decrease in oceanic areal type is observed. On the other hand, in the conditions of dry pontic continental climate in Vojvodina province, highest number of continental species and those with xeric affinities (mostly Mediterranean) is documented, compared to the other parts of the country. The analysis of geoelements among bryophytes within different regions of Serbia, gives a new insight from the conservational point of view. Target is not given on bryophyte species only, but on unique bryophyte assemblages of specific origin, composition and/or biogeographical spectra present in certain areas as well. Apart from data on species presence, this is a valuable input for preparing of bryologically important areas network of Serbia (IBrAS).

**KEYWORDS:** geoelement, distribution, ecological divergence

Poster presentation 12 10 02

### IS THERE ANY PATTERN IN BRYOPHYTE ECOLOGICAL AND LIFE HISTORY TRAITS AT HEAVY METAL LOADED SITES? CASE STUDIES FROM SLOVAKIA

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Two spoil heaps in Central Slovakia, namely Malachov – Veľká studňa and Lubietová – Podlipa, were chosen to study spontaneously established bryophytes. The spoil heaps were selected so that both are heavy metal loaded but with different toxic elements (Veľká studňa with mercury, and Podlipa with mainly copper) and that both have similar climatic conditions. This is important for distinguishing between heavy metal influence on bryophyte assemblages. Interestingly, the ratio of liverworts and mosses was nearly the same (10:90%). At both sites

the most abundant were hypnacean species, followed by bryalean ones. Among bryophytes recorded, perennial way of living was very common at both sites (ca. 90%). The studied sites also shared the life form frequency where the most abundant were bryophytes growing vertically with little or no branching (turfs). Dominant life strategies among bryophytes at both heaps were competitive perennials and perennials, followed by colonists. Most of the species were adapted to medium to open light and cooler conditions, according to abiotic factor indices. A moderate continentality (drought support) among species recorded was the most common but species adapted to spectra of wet conditions were also indicative. At Podlipa there was a significant number of rather acidophile species while this was absent at Velká studňa where many records of basiphile bryophytes were made. At both sites, many species preferred moderate acidity. When nitrogen index is taken into account, mostly eutrophic species are present at both sites. Interestingly, according to heavy metal index, most of the species at both heaps occur rarely on substrates with moderate or high concentrations of heavy metals implying that the bryophyte assemblages are living in suboptimal environment. There is a rather small percentage of species that are frequent and often abundant on substrates with moderate or high concentrations of heavy metals, sometimes occurring as dominants over large areas, but also frequent in other habitats. The results obtained clearly showed similar patterns in the bryophyte composition at both studied sites. These organisms are indicative to the site resources and some species among the recorded ones can be metal-tolerant and even regarded as potential indicators for certain environment parameters.

**ACKNOWLEDGEMENTS:** The study was supported by the Scientific Grant Agency of the Slovak Republic under Grant VEGA 2/0040/17.

**KEYWORDS:** bryophytes, traits, spoil heaps, Malachov, Lubietová, Slovakia

Poster presentation 13 10 09

### IS THERE A TOLERANCE TO SALT AMONG NON-HALOPHYTIC POLYTRICHACEOUS BRYOPHYTES?

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A huge group of land plants, namely bryophytes, can be found in all ecosystem types except salt water. Though, there are rare bryophyte representatives that can survive in salty harsh environment. Also, it was shown that the tolerance to salt stress is present within the non-halophytic model moss *Physcomitrella*

*patens*. With aim to test if this is common feature within plant subkingdom of bryophytes, we chose two related non-halophytic moss species, namely *Polytrichum formosum* (two genotypes) and sibling species *Atrichum undulatum*, both inhabit mesophytic forest floors. The *in vitro* cultures were established and gametophores were transferred to short and long salt stress exposure with aim to test their salt tolerance. The salt was added to the medium in range (0-500 mM), and the following developmental and biochemical parameters were measured: survival, secondary protonemal diameter development, index of multiplication, tocopherol, chlorophylls, xanthophylls and carotenoids. The results obtained clearly showed that there is a certain extent of tolerance to NaCl (present in substrate) in both species. *Polytrichum formosum* could slightly better adapt to salt stress compared to *Atrichum undulatum*. However, there was significant physiological difference in two *P. formosum* accessions suggesting better tolerance in Serbian compared to German genotype. Short and long exposure of both species brings expected results only to some extent, having in mind that biochemical parameters (i.e. chlorophyll and tocopherol content) showed also interesting patterns of different tolerance within two genotypes of *P. formosum*. In all tested accessions, salt tolerance is present. It is species specific, genotype specific and depends on the concentration and time of exposure. Short exposure to salt is not necessarily easier to adapt by tested mosses, but it is easier to survive.

**KEYWORDS:** mosses, salt stress, adaptation, biochemistry

Poster presentation 14 10 08

### EXPERIMENTAL APPROACHES ON BIOTIC RELATIONSHIPS AMONG BRYOPHYTES AND FUNGI IN THE CONTROLLED CONDITIONS

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Bryophytes and fungi share habitats and compete for the same environmental resources. During history they co-evolve and developed complex of relationships like pathogene, parasitism, saprobe or commensalism. Additional effects can be described as documented endobiotic life strategy of fungi within bryophyte species, effects of fungi on endoreduplication i.e. endopolyploidy in mosses or allelopathy. While some fungi have no clear influence on bryophytes other like bryophytic fungi are completely dependent on bryophytes as hosts. Thus, many interesting relationships between these two groups have been already noticed. With aim to study biotic relationships between bryophytes and fungi, we chose six fruit

pathogene fungal species (namely *Botryosphaeria dothidea*, *Botrytis cinerea*, *Phomopsis viticola*, *Colletotrichum acutatum*, *Monilinia laxa* and *Calosphaeria* sp.) and additional eight bryophyte species (namely mosses *Cinclidotus fontinaloides*, *Anomodon viticulosus*, *Thuidium tamarsicinum*, *Eurhynchium striatum*, *Isothecium alopecuroides* and *Polytrichum formosum* and liverworts *Porella platyphylla* and *Scapania nemorea*). The experimental approach was designed to test the effects of various extract types (ethanol and methanol) of each bryophyte to each fungal species by the disk diffusion methods. The network of relationships was studied and it can be concluded that some bryophytes have negative effects to some fungal species, some are indifferent while there was no positive interaction detected. The results obtained clearly showed that some bryophyte species show high resistance to fungal counterparts (*Scapania nemorea*, *Isothecium myosuroides* and *Anomodon viticulosus*) while some other rather evolve so to have indifferent or tolerant relationship to selected fungal species. Fungi were either tolerant or sensitive to the presence of bryophytes. This specific relationship where fungi express sensitivity on selected bryophyte species can be further studied with an aim to develop the fundamental bases for biotechnological approach of biofungicide production and application *in vivo*. For the huge biomass production of bryophyte material, *in vitro* establishment of the bryophyte species expressing the most suppressing fungal growth were done. At present, the growth optimization in the controlled conditions of these bryophyte species are in progress.

**KEYWORDS:** mosses, liverworts, fungi, relationships, biofungicide

Poster presentation 15 10 03

### WOOD DECAYING FUNGI IN THE ÇORUM PROVINCE FORESTS (TURKEY)

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Due to its location, flora and climate conditions, Turkey is very rich in terms of diversity of wild growing macrofungi species. Because wood decaying fungi can degrade cellulose and lignin in nature, they have an important role in the cycle of organic matter in the natural ecosystems, especially forests. The aim of this study was to determine the presence and distribution of wood decaying fungi in Çorum province. The city is in central Turkey, inland from the Black Sea coast. Çorum is on a high plateau with a typical inland climate of with light rain in spring and autumn, hot, dry summers and cold, snowy winters. Çorum province and around have attractive mountainous countryside, and this area does not protect by any establishment. During the field trips, in Çorum province between the years 2011 and 2015, fruiting bodies were collected, and photographed in their natural habitats. The ma-

ture specimens collected during the periods were determined by using the related literature and the dried specimens were deposited into fungarium for future use. Macrofungi samples are kept in the Fungarium of Selçuk University Directorate of Fungi Application and Research Center. As a result, 71 taxa were determined within two phyla *Ascomycota* and *Basidiomycota*. Among all documented 18 species belong to phylum *Ascomycota*, while the rest of them belong to *Basidiomycota* division. Consequently, in phyla *Ascomycota*, 18 species belonging to 9 families and 16 genera were identified. Similarly, in *Basidiomycota* 53 species belonging to 22 families and 37 genera were identified. Fungi, grown on wood, cause brown and white rot. Usually leaving the lighter-colored cellulose behind, white rot fungi decompose the lignin in wood. On the other hand, cellulose and hemicellulose that form the wood structure are broken down by brown rot fungi. According to the literature survey, 36 fungi were found to cause white rot, 30 fungi were found to cause brown rot in the ecosystems of the forests.

**KEYWORDS:** systematic, macrofungi, brown and white rot, tree, Çorum, Turkey

Poster presentation 16 10 14

### CURRENT KNOWLEDGE ON DIVERSITY OF HYPOGEIC MACROFUNGI OF SERBIA WITH THE FOCUS ON WESTERN SERBIA

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The serious collection of hypogeous macrofungi in Serbia has begun in the beginning of 1990's when MM started to form the collection of the dry exsiccates. Since 1998 these exsiccates were examined and also additional collected and determined by other authors of this contribution. The investigated territory is a small portion of potential habitats for such fungi, mostly because their detection was in a great deal a side effect of truffle collection and therefore limited to truffle habitats. So far 14 different species of real truffles (*Tuber* sp.) were detected and determinations confirmed by molecular phylogeny methods. Other than that, we have detected and determined 45 species belonging to 20 genera (Class Mucoromycota (1), Ascomycota (10) and Basidiomycota (9)) of fungi not belonging to the genus *Tuber* sp. In this contribution we present the detected species, and especially concentrate on those collected in Western Serbia and their habitats. All collections originating from Western Serbia are preserved in private herbarium of AG. The so far detected diversity of hypogeous fungi in Serbia is not high comparing to other South European regions, but is also far from being well explored and certainly

needs further investigation.

**ACKNOWLEDGEMENTS:** Some of these results originate from the investigations undertaken during EUREKA E!3835 EURO-AGRI- CULTUBER "Improvement of truffle cultivation via novel quality control, soil analysis and inoculation methods", while great amount of data originate from the private financed investigation. ŽM and NV are also financed by project III43010 financed by Ministry of Education, Science and Technological Development of Serbia.

**KEYWORDS:** hypogeous, macrofungi, truffles, diversity

Poster presentation 17 10 13

### ARBUSCULAR MYCORRHIZAL FUNGI IN SELECTED ECOSYSTEMS IN SERBIA

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Mycorrhiza is an obligatory symbiosis between fungi and 99% of terrestrial plant species, majority of which form the so called arbuscular mycorrhiza (AM). However, one of the European regions with highest plant diversity rates, Balkan Peninsula, has never been explored for diversity of AM fungi (AMF). AM has previously been considered dominant type of mycorrhiza in tropics and communities formed by herbaceous plants, while northern temporal and boreal forest ecosystems have been explored for AMF diversity only recently. Unexpectedly, these forests, thought to be dominated by ectomycorrhiza, appeared to host rich AMF communities as well. In South European regions such topics have rarely been addressed. In order to start investigation of AMF diversity in Serbia, we have applied two approaches: we set three season-based experiments in different ecosystems aiming to explore in depth the influence of dominant ecological factors on AMF communities; and we have sampled soil in 20 different natural and agricultural locations all over the country in order to get a broad idea on AMF diversity. For approaching AMF communities we have applied comparative 18S DNA NGS based analyses of sampled roots or soil and spore morphology based analyses from soil samples taken from the rhizosphere. All investigated soils have been analyzed as well. Here we present the current state of the results archived by spore morphology analyses. From experiments we present data on 46 species seasonal distribution and abundance, while from country overview we present the concept of distribution and so far detected 51 species and several undescribed taxons.

Only on these preliminary data, it is clear that Serbia hosts very high diversity of AMF.

**ACKNOWLEDGEMENTS:** This research has been financed by Swiss National Scientific Foundation through SCOPES initiative project IZ73Z0\_152740. NV and ZM are also financed by project III43010 financed by Ministry of Education, Science and Technological Development

**KEYWORDS:** mycorrhiza, arbuscular, fungi, diversity, ecology

Poster presentation 18 10 27

### FIRST REPORT OF SOME ENDEMIC PLANT SPECIES IN CROATIA AS NEW HOST PLANTS FOR PHYTOPATHOGENIC FUNGI

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Phytopathogenic fungi cause various diseases or phytomyces on plants in agroecosystems, but also on plants in natural ecosystems. The Flora of Croatia is very rich in endemic plant species (384 taxa), but chorology and epidemiology of phytopathogenic fungi on endemic plant species in Croatia is not investigated well enough. Incidence of phytopathogenic fungi can have a negative impact on endemic plant species, especially on stenoendemic species. In order to determine occurrence of phytopathogenic fungi and etiology of phytomyces on endemic plants in Croatia, appearance of disease symptoms on some endemic plant taxa in national parks (Velebit and Paklenica), nature parks (Biokovo and Žumberačko-Samoborsko gorje) and Botanical garden in Zagreb was monitored. Samples of endemic plant taxa with disease symptoms from genera: *Aguilegia*, *Berberis*, *Campanula*, *Dianthus*, *Helleborus*, *Iris*, *Lilium*, *Primula* and *Sibiraea* were collected and analyzed in mycologically laboratory. Identification of phytopathogenic fungal species was performed on the basis of their morphological characteristics (spores and spore-bearing structure). Ten phytopathogenic fungal species were determined on sixteen endemic plant taxa. Fungus *Erysiphe aguilegiae* DC. was determined on *Aguilegia kitaibellii*; fungus *Puccinia brachypodii* Otth was determined on *Berberis croatica*; fungus *Coleosporium campanulae* (Pers.) Tul. was determined on *Campanula fenestrellata* and *C. fenestrellata* ssp. *istiraca*; fungus *Mycosphaerella dianthi* (Burt) Jorst. was determined on *Dianthus giganteus* ssp. *croaticus*; fungus *Microsphaeropsis hellebori* (Cooke&Masse) Aa was determined on *Helleborus atrorubens*, *H. multifidus* and *H. niger* ssp. *macranthus*; fungus *Cladosporium iridis* (Fautrey& Roum.) de Vries was determined on *Iris adriatica*, *I. croatica*, *I. illyrica* and *I. pseudopallida*; fungus *Puccinia iridis* Wallr was determined on *Iris croatica* and *I. pseudopallida*; fungus *Botrytis* sp. was determined on *Lilium martagon* ssp. *cattaniae*; fungus *Septoria primulicola* Rostr. was determined on *Primula kitaibelina*

and fungus *Monilina* sp. on *Sibiraea altaensis* spp. *croatica*. All mentioned endemic plant species (except *Aguilegia kitaibelli*) are new host plant for these phytopathogenic fungi.

**KEYWORDS:** Croatia, endemic plants, phytopathogenic fungi, phytomyces

Poster presentation 19 10 16

### SCHIZOPHYLLUM COMMUNE FR. 1815 FROM SUBMERGED CULTURES – INHIBITION OF ACHE ENZYME AND ANTIOXIDATIVE ACTIVITY

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*Schizophyllum commune*, split gill mushroom, is a widely distributed wood-decaying basidiomycete that has been reported for its health promoting and medical benefits. Due to more people worldwide are suffering from Alzheimer's disease (AD), a chronic neurodegenerative disorder that usually starts slowly and worsens over time, with no treatments to stop or reverse its progression, the solution is sought in nature while acetylcholinesterase inhibitors (AChEIs) became an attractive research subject. In recent years fungi have been recognized as producers of AChEIs, agents that inhibit the enzyme AChE from breaking down acetylcholine, thereby increasing both the level and duration of action of the neurotransmitter acetylcholine (ACh). Since fungi produce a variety of metabolites in submerged cultivation of *in vitro* grown mycelia (M) and fermentation broths (F), the aim of the study was to examine and compare the AChE inhibitory and antioxidative activity of ethanol extracts (80%) both from M and F of two different strains of *Sch. commune* originated from Serbia (SRB) and Italy (IT) during 28 days submerged cultivation. The highest anti-DPPH radical activity was achieved for F extracts being increased during cultivation. The IT strain *Sch. commune* F (12,55µg/mL) showed the highest activity after 28 days, while SRB strain obtained the highest ferric reducing ability after 21 days (228,46 mg eq. AA/g d.w.). The AChE activity was determined using Ellman's method and presented as an IC<sub>50</sub> (50 % inhibitory concentration) value. All screened extracts exhibited AChE activity, with isolate IT being more active. Furthermore, the results showed that after 14 days cultivation the AChE activity was the highest (for F: IC<sub>50</sub> 1.8 mg/ml SRB, IC<sub>50</sub> 0.8 IT mg/ml; for M: IC<sub>50</sub> 2.7 mg/ml, SRB IC<sub>50</sub> 1.7 mg/ml IT), at the same time showing that all F extracts were more active than M extracts. According to the growth curves of both fungal strains, we assumed that different metabolites,

both primary and secondary could inhibit the AChE. In summary, this fungal species is well worth for further research, since both isolates represent a promising resource of natural AChE inhibitors that might improve palliative therapy of AD in the future.

**KEYWORDS:** *Schizophyllum commune*, submerged cultivation, AChE inhibitors, Alzheimer's disease, antioxidants

Poster presentation 20 10 25

### CONTRIBUTION TO THE LICHEN FLORA OF MACEDONIA AND ALBANIA

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Macedonia and Albania are small European countries in the southwest of the Balkan Peninsula only poorly understood in a field of lichenology. This is true especially for Albania due to its isolation during last century and lack of lichenologist in the country. Both examined countries are mountainous and Albania is also a seaside country. This poster presents new records of lichens from these areas compared to countries respective checklists of species. For lichen identification, traditional techniques such as microscopy, binocular loupe and spot tests were used. Selected localities in Macedonia (Treskavec monastery's hill, Baba Mountain – Pelister peak, Galičica National Park) and in Albania (National Park Llogara, Maraç near Vau i Dejes, village Mezi, valley Valbona in Prokletije mountains and pass near village Leskovik) were studied during a field excursion in May 2018. Total of 68 species were recorded. For Macedonia, we present 6 newly listed species, respectively 21 new species for Albania. Valuable addition is also discovery of lichenicolous fungi new to Albania – *Plectocarpon lichenum*. *Biatora efflorescens*, *Cladonia diversa*, *Cladonia grayi*, *Lecidea berengeriana*, *Umbilicaria spodochoa* are not only new to Macedonia or Albania but rare in a whole Balkan region.

**KEYWORDS:** lichen, Macedonia, Albania, taxonomy, lichenicolous, *Plectocarpon*

Poster presentation 21 10 31

### SOME INTERESTING RECORDS OF LICHENICOLOUS FUNGI LIVING ON *THAMNOLIA VERMICULARIS* IN SERBIA

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Despite of the intensive floristical investigations of the Balkan Peninsula in the last decades, there are many undiscovered or unpublished taxa, especially among microscopical fungi invading lichen thalli. In higher altitudes in the alpine regions cryptogam organisms like lichens are conspicuous both in diversity and biomass. The estimated number of lichenicolous fungi from Serbia is much higher than the currently known species number (Savić & Tibell 2006; Bilovitz & Mayrhofer 2008). The number of thamnicolous species is more than 20 worldwide (Zhurbenko 2012). The aim of our research is to contribute to the number and location of lichenicolous fungi from Serbia and also from the Balkan Peninsula. The study is based on collections and fieldworks during the years 2000–2016. The collected material is deposited in the Lichen Collection of the Hungarian Natural History Museum, Budapest (BP). *Thamnolia vermicularis* is an easily identified fruticose lichen species by its 10–40 mm long, ivory-white tubular thallus, with prostrate or erect fruiting bodies. It has widespread arctic-alpine distribution with two distinct chemotypes. Our material was UV+ orange under ultraviolet light (365 nm). Specimens were investigated from 3 high mountain localities of Serbia (Kopaonik Mts, Stara Planina Mts, Suva Planina) using light microscopical methods and hand made sections. Our results show that several lichenicolous taxa seem to be new for the Midžor region like *Cercidospora*, *Cornutispora*, *Endococcus* and *Thamnogalla* spp.

**ACKNOWLEDGEMENTS:** Our research was supported by the projects Hungarian Scientific Research Fund OTKA K81232 and the National Research, Development and Innovation Fund NKFI K124341.

**KEYWORDS:** lichens, lichenicolous fungi, *Thamnolia vermicularis*, Serbia

Poster presentation 22 10 21

### SECONDARY METABOLITES OF LICHENS AS BIOTIC FACTORS INDUCING CHANGES IN THE PHYSIOLOGY OF MOSSES

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Mosses and lichens in nature share similar habitats, but principle of this cohabitation is not well known to date. Lichens try to disadvantage their competitors through releasing allelochemicals into an environment, where neighboring mosses and other organisms grow. These allelochemicals represent secondary metabolites of lichens. This work aimed to expand the knowledge on allelopathic effects of lichen secondary metabolites on bryophyte physiology. In this study, we used extract isolated from lichen *Pseudevernia furfuracea* containing secondary metabolites atranorin, chloratranorin and physodic acid as a major compounds. This quantitative identification of lichen secondary compounds was carried out by HPLC (High Performance Liquid Chromatography). To observe the allelopathic effect of these metabolites, selected species of mosses *Physcomitrella patens* and *Pohlia drummondii* were cultivated under aseptic condition for 5 weeks on sterilized glass fiber disks containing these compounds. The natural extract of secondary metabolites was tested in two selected doses (0.01% and 0.1%). In the experimental work we spectrophotometrically analyzed the content of photosynthetic pigments, carotenoids, anthocyanins and soluble proteins. Experimentally, we found that in the case of *Physcomitrella patens* the application of secondary metabolites did not significantly affect the content of pigments, carotenoids or anthocyanins. In the case of *Pohlia drummondii*, there was a statistically significant difference in the assimilation pigment content of carotenoids at both tested concentrations. In our study, there was a statistically significant difference in the soluble proteins content of both tested mosses after application of the highest concentration of secondary metabolites. From these results, we conclude that lichen secondary metabolites act as potential allelochemicals which influence changes in the physiology of mosses.

**KEYWORDS:** lichens, secondary metabolites, mosses, allelopathy

Poster presentation 23 10 29

### ANTIMICROBIAL ACTIVITY OF SILVER NANOPARTICLES PREPARED BY LICHEN-BASED MECHANOCHEMICAL SYNTHESIS

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During the last decades, microorganisms have developed resistance against traditional antibiotics. Therefore, pharmaceutical industry tries to find alternative therapeutic agents and new methods of their synthesis. The natural products exhibiting antibacterial activity are particularly attractive. Silver nanoparticles are known for their antibacterial potential. There are many synthetic methods for their preparation among which the green synthesis in which natural plant material is used as a reduction agent is still gaining more popularity. A specific alternative of green synthesis is bio-mechanochemical synthesis, in which the nanoparticles are synthesized in solid state by directly co-milling of plant material with silver precursor. Vast amount of various plant extracts is being applied for Ag NPs preparation these days. However, the reports on the lichen-based synthesis are very scarce. Moreover, lichens also exhibit antibacterial activity. Numerous recent review papers demonstrated that secondary metabolites of lichens are responsible for this action. In the present study, two lichen species *Pseudevernia furfuracea* and *Lobaria pulmonaria* were used for bio-mechanochemical synthesis of silver nanoparticles by ball milling with AgNO<sub>3</sub> for two hours. Their antibacterial activity was tested on 6 bacterial strains and compared with that of pure lichen extracts. The samples containing silver nanoparticles had significantly higher effect on both gram negative and gram positive bacterial strains. For *P. furfuracea* also water extract without Ag NPs was effective against two bacterial strains (*Staphylococcus aureus* and *Listeria monocytogenes*). Pure *L. pulmonaria* extract did not show any antimicrobial activity.

**KEYWORDS:** lichens, green synthesis, mechanochemistry, nanoparticles, antimicrobial activity

Poster presentation 24 10 19

### CHEMICAL CHARACTERIZATION AND ANTIBACTERIAL ACTIVITY OF THE EXTRACTS OF THREE LIVERWORT SPECIES

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Liverworts contain mono-, sesqui- and diterpenoids and aromatic compounds [bibenzyls, bis-bibenzyls, naphthalenes, phthalides, isocoumarins, cinnamates, benzoates etc.] which constitute the oil bodies (specific cellular organelles found in almost all leafy liverworts and thalloid liverworts). Chemical analysis and antibacterial properties of methanol extracts of three thalloid Hepatics (*Marchantia polymorpha*, *Conocephalum conicum* and *Pellia endivifolia*) are investigated. <sup>1</sup>H NMR spectrometry was used for analyzing the main group of compounds in methanol extracts of three liverwort species. Two assays have been used for testing antimicrobial properties of the extracts (well diffusion and microdilution method) on selected Gram (–) and Gram (+) bacteria. <sup>1</sup>H NMR analysis of the methanol extract of *P. endivifolia* revealed bisbibenzyls, terpenes and sugars as dominant compounds. In *M. polymorpha* extract, marchantin A was found to be by far dominant bis-bibenzyl (comparing to marchantin A standard). In *C. conicum* methanol extract bis-bibenzyls are absent, while terpenes and sugars are present. Results obtained from antibacterial activity showed that methanol extracts from all three species, as well as standard of marchantin A (pure compound isolated from *M. polymorpha*) don't inhibit Gram (–) bacteria tested (*Xanthomonas arboricola*, *Escherichia coli* ATCC 25922, *Pseudomonas aeruginosa* ATCC 27853 and *Pseudomonas syringae* CFBP2473). Gram (+) bacteria (*Staphylococcus aureus* ATCC 25923, *Clavibacter michiganensis*, *Listeria monocytogenes* ATCC 19111, *Bacillus subtilis* ATCC 6633) have been sensitive to all extracts tested except to *C. conicum* methanol extract. The strongest effect showed marchantin A which inhibit growth of *S. aureus* at concentration of 0.062 mg/ml and inhibition zone of 11 mm.

**KEYWORDS:** liverworts, marchantin A, NMR spectroscopy, antimicrobial activity

Poster presentation 25 10 07

### DIVERSITY OF STONEWORTS (CHAROPHYTA) IN THE SNR “GORNJE PODUNAVLJE“

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The Monoštor marsh is a part of the Special Nature Reserve “Gornje Podunavlje“, which makes a majority of recently proclaimed UNESCO Biosphere Reserve “Bačko Podunavlje“. This unique marshland area in the Danube floodplain is considered to be one of the last flora and fauna refuges in the region. The aim of our study was to investigate diversity and distribution of the charophytes in five selected ponds in the Monoštor marsh. The selected ponds were: Semenjača, Šarkanj, Široki rit, Sakajtaš and Ribolov. The pond Široki rit is the chronologically last in a series of three revitalized ponds; formerly Semenjača and Šarkanj were revitalized. The ponds Ribolov and Sakajtaš still weren't exposed to the revitalization processes. The ponds were sampled monthly, in the period May – September 2016. Samples were collected with the aid of the specifically constructed tool and the shafts, from the coast. The stonewort samples were identified using STEMI DV4 stereomicroscope and Nikon YS100 microscope and standard literature. Charophyta representatives in macrophyte communities were detected in only three, out of five selected ponds - Ribolov, Sakajtaš and Široki rit pond. In total, eight species of charophytes were detected: *Chara contraria*, *C. globularis*, *C. tenuispina*, *C. vulgaris*, *Nitella confervacea*, *N. gracilis*, *N. syncarpa* and *Tolypella prolifera*. The highest diversity of charophytes was noted in Široki rit pond and considerably lower in Ribolov pond, while only one species - *C. contraria* was detected in Sakajtaš pond. This undoubtedly points to the stoneworts success in newly established conditions during revitalization process in Široki rit pond, probably because promoted oospore germination process due to excavation. All the stonewort species detected in this survey were previously recorded in Vojvodina, i.e. Serbia, still *Chara tenuispina*, *Nitella confervacea* and *Nitella gracilis* so far haven't been found in the territory of the Monoštor marsh. Thus, these records should be considered new field records, which are valuable data since IUCN threat statuses of all three species is critically endangered in Serbia, but also endangered in Balkan Peninsula.

**KEYWORDS:** stoneworts, shallow ponds, revitalization, *Chara*, *Nitella*, *Tolypella*

Poster presentation 26 10 32

### CHARACTERISATION OF NOSTOCALES CYANOBACTERIA STRAINS ISOLATED FROM FRESHWATERS OF GREECE

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Cyanobacteria are a diverse group of microorganisms that inhabit a wide range of ecological niches and are well known for their toxic secondary metabolite production, which can be a threat for human and environmental health during and after water-blooms. The characterization of the bloom communities remains challenging because the taxonomy of certain cyanobacterial genera has not yet been resolved, especially within some potentially toxic Nostocales taxa, due to the co-occurrence of several different morphotypes. The comparison of morphological and genetic data is sometimes hindered by the lack of cultures of several cyanobacterial morphospecies and inadequate morphological data of sequenced strains. In this context, we studied 11 isolates belonging to *Anabaena*, *Anabaenopsis*, *Chroococciopsis*, *Cylindrospermopsis*, *Dolichospermum*, *Hapalosiphon*, *Nostoc*, and *Sphaerospermopsis* genera from freshwater ecosystems of Greece, deposited in Thessaloniki Aristotle University Microalgae and Cyanobacteria culture collection (TAU-MAC / <http://cyanobacteria.myspecies.info/>), with respect to their morphological and genetic characteristics. The strains were morphologically identified during isolation, at least down to the genus level, and preserved in liquid cultures of BG11 medium under suitable light and temperature conditions. We extracted the whole genomic DNA and amplified the 16S rRNA and phycocyanin operon subunits (cpcBA) using Polymerase Chain Reaction. We sequenced the products, compared the sequences of all strains with those deposited in public databases and performed phylogenetic analysis. Phylogenetic analysis indicated that strain TAU-MAC 0115, morphologically determined as *Hapalosiphon*, clustered with a *Scytonema* cf. *crispum* strain, but within Hapalosiphonaceae clade, deserving further investigation. *Nostoc* sp. TAU-MAC 0515, isolated from a rice field, formed a clade with *Nostoc oryzae* strains, whilst *Nostoc* sp. TAU-MAC 0215 clustered with other *Nostoc* strains. Interestingly, *Dolichospermum* sp. TAU-MAC 1714 formed a separate clade within *Nostoc*. Our findings are in line with the phylogenetic diversity often reported in Nostocales cyanobacteria, whilst we highlight that the high degree of morphological complexity in some Nostocales genera is in congruence with their phylogenetic heterogeneity.

**KEYWORDS:** Nostocales, Phylogeny, Greece, *Nostoc*, *Hapalosiphon*, *Scytonema*

Poster presentation 27 10 06

### ALLELOPATHIC ACTIVITY OF MYRIOPHYLLUM SPICATUM L. ON NATURAL PHYTOPLANKTON ASSEMBLAGES

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Worldwide and comprehensive eutrophication of the water bodies and consequential occurrence of toxic algal blooms became one of the most serious environmental problems. Living in the same water habitat with a limited amount of resources, aquatic macrophytes and microalgae compete for nutrients and especially light, that is considered as limiting factor. During the long-time coevolution, each of those groups have developed numerous defensive strategies against other photosynthetic organisms competing for light. Allelopathic inhibition is considered to be an effective macrophyte competitive strategy against algae that can bloom and thus significantly decrease an amount of light that reaches to the macrophytes. The objective of this research was to investigate the allelopathic activity of the different concentrations of the *Myriophyllum spicatum* L. extracts on the structure and dynamics of natural phytoplankton assemblages in *ex situ* conditions. Three different amounts of dried and powdered plant material were solved into the equal volume of 40% ethanol, making three concentrations (A (1g/50ml), B (5g/50ml) and C (25g/50ml)) of *M. spicatum* extract. 1 ml of each extract was added into three experimental flasks containing phytoplankton samples collected from the lake and precultivated on the BG-11 medium. Sub-sample of microalgae suspension was taken from each flask four times during the experiment: before extract application, 4, 8 and 24 h after adding extracts. Qualitative and quantitative analyses of the taken sub-samples were done. The obtained results showed that all of three concentrations of the *M. spicatum* extracts showed the inhibitory effect to a certain extent. The B extract showed the strongest inhibitory effect on the total algal biomass, 4 h after extract adding, which is confirmed by the redundancy analysis of the experiment results. The inhibitory effect of extracts is evident within all recorded algal phyla. Phylum Cyanobacteria is found to be the most susceptible to applied extracts compared with phyla Chlorophyta and Bacillariophyta. Those results give the opportunity for more specific research of the potential allelopathic control of harmful algal blooms and sustainable management and recovery of water bodies as an ultimate goal.

**KEYWORDS:** Allelopathy, antialgal activity, extracts, *Myriophyllum spicatum* L., natural phytoplankton assemblages

Poster presentation 28 10 20

### KEY SPECIES THAT PRODUCED PHYTOPLANKTON BLOOMS IN THE ROMANIAN BLACK SEA WATERS DURING 2001-2017

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This paper represents an overview of the most common microalgae producing bloom events in the Romanian Black Sea waters during 2001-2017. The objective was to observe the proportion of the main algal groups which dominated the phytoplankton community and the physico-chemical conditions in which these events occurred. Consequently, we analysed phytoplankton and chemistry data sets of the National Institute for Marine Research and Development from the Romanian Black Sea coastal waters between 2001 and 2017, to overview the most important algal blooms. The biological samples collected using Nansen bottles from standard depths (0, 10, 20, 40m) were preserved with 4% formaldehyde seawater buffered solution and processed by the sedimentation method. We did the taxonomic identification of species and counting of cells in a 0.1 ml fraction of each sample with Olympus inverted microscope, using a 40x objective lens for nanoplankton (less than 15–20 microns) and 10x or 20x for larger cells. Due to available equipment in the earlier years, temperature and salinity were measured with a reversible thermometer and the volumetric method, and recently by CTD (model YSI Cast Away); nutrient samples were determined according to the standard methods for seawater analysis (Grasshof, 1999). Within 2001-2017, 25 species of microalgae bloomed on the Romanian Black Sea coastal waters. The diatoms dominated the community in most of the years representing between 56-99% of the phytoplankton annual average density. For the studied period we found only two significant blooming events, one in 2003 (over 50·10<sup>6</sup> cells/litre) and the other in 2010 (39.5·10<sup>6</sup> cells/litre). From the abiotic elements, the salinity and temperature reached high variability, but we found constant oxygen oversaturation confirming the blooms and high nutrients concentrations as their cause.

**KEYWORDS:** microalgae, Black sea, phytoplankton, diatoms

Poster presentation 29 10 12

### THE RELATIONSHIP BETWEEN EPILITHIC DIATOM COMMUNITIES AND ECOLOGICAL STATUS OF THE RADOVANSKA RIVER (SERBIA)

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Rivers and streams are one of the most endangered ecosystems in the world, so it is necessary to assess the ecological status and continuous monitoring of these ecosystems. Anthropogenic effects, but also many inorganic processes such as erosion, degrade surface waters. This affects ecological integrity and the functioning of freshwater ecosystems, and thus disables their use. This has resulted in the creation and use of diatom indices worldwide based on diatom communities. Assessment of the ecological status of the Radovanska River based on the epilithic diatom communities and diatom indices as well as consideration of the efficiency of their use in the assessment of the ecological status of the rivers in Serbia were the main aims of this work. Epilithic diatoms were sampled at 5 sampling sites along the Radovanska River, during 2011-2012 by scraping from stones with a toothbrush. The relative abundance of taxa was determined by counting 400 valves on each slide after diatom frustules cleaned using strong acids and making permanent slides. Values of diatom indices were obtained using the OMNIDIA software. Canonical correspondence analysis (CCA) grouped the diatom taxa into three main clusters based on the physico-chemical parameters. The first cluster included taxa significantly positively correlated with nitrates. These were: *Achnantheidium catenatum*, *Coconeis pediculus*, *Gomphonema exilissimum*, *Luticola mutica*, *Nitzschia archibaldi*, *Staurosira mutabilis*, *Surirella angusta*. The second cluster (*Adlafia minuscula*, *Mayamaea permitis* and *Navicula antonii*) involved taxa which showed positive correlation with oxygen. *Cymbella compacta*, *C. excisa*, *C. parva*, *Encyonema minutum* and *Gyrosigma acuminatum* made up the third cluster and were significantly positively correlated with total phosphorus. Most indices indicated good and high ecological status of the Radovanska River. In Serbia, two diatom indices (Specific Pollution Index-IPS and European Economic Community Index-CEE) are a mandatory parameter in assessing the ecological status/potential of surface waters. According to our legislation and based on phytobenthos, the ecological status of the Radovanska River is assessed as high.

**KEYWORDS:** diatom indices, epilithic diatom communities, bio-monitoring, ecological status

Poster presentation 30 10 23

### CHANGES IN THE DIATOM COMMUNITY STRUCTURE AND IMPACT ON WATER QUALITY FROM 2005-2017 - GREAT LAKE (LURË NATIONAL PARK, ALBANIA)

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Great Lake is the largest of the 6 lakes situated in the eastern part of Lura Mountain in the Lurë National Park. Samples were collected in July 2005, June and August 2013 and August 2017. In 2005 and August 2013 epiphytic samples were collected from one locality, while in Jun 2013 was collected epilithic sample. However, in August 2017 no macrophytes were found which caused the collection of epilithic samples from 6 localities (LM1-LM6) around the lake with the aim of establishing ecological status. After laboratory analysis 52 taxa were identified in July 2005, 67 in June 2013 and 111 in August 2013. In August 2017 the number of identified taxa was higher, in total 126 taxa. Dominant taxa in July 2005 were *Achnantheidium minutissimum* (13.01%), *Diatoma vulgare* (13.84%) and *Meridion circulare* (11.76%). *A. minutissimum* was also dominant in 2013 (22.26% in June and 21.48% in August). In 2017, dominant taxa differed among localities: LM1 *Cyclotella cretica* var. *cyclopuncta* (29.38%), LM2 and LM3 *Encyonopsis microcephala* (21% at both sites), LM4 *Cyclotella cretica* var. *cyclopuncta* (17.41%) and *Encyonopsis krammeri* (18.91%), LM5 and LM6 *A. minutissimum* (27.79% and 30.47%, respectively). The genera *Diatoma*, *Epithemia*, *Fragilaria* and *Surirella* were not recorded in 2017, contrary to previous years. The ecological status of the Great Lake was assessed by using diatom indices. Most European countries have established lake assessment systems based on diatoms. However, the Albanian National Monitoring System does not include biological parameters and therefore no officially accepted diatom index is in place. We compared the values of IPS (Indice de Polluo-sensibilité) and TDIL (Trophic Diatom Index for lakes) indices using boundary values given by the authors. Values of IPS index indicated very good water quality in all years at all sites, while the TDIL indicated moderate water quality in 2005 and 2013 and good water quality in 2017. The reason for this deviation probably is the low relative abundance of indicator species used to calculate TDIL index. The TDIL was developed 2007 in Hungary, and it includes significantly fewer indicator taxa than the IPS. More comprehensive ecological investigations of lakes in Albania are necessary to be able to fulfill the demands of the Water Framework Directive.

**KEYWORDS:** diatoms, Lurë National Park, Great Lake, IPS, TDIL

Poster presentation 31 10 11

### SMALL RIVERS – SOURCE OF HIGH DIATOM DIVERSITY: CASE STUDY OF THE CRNICA RIVER (SERBIA)

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Diatoms (Bacillariophyta) are widespread in different types of aquatic ecosystems, often with high abundance. They are one of the most important producer and ecologically important groups in rivers and streams. Therefore, represent significant elements in biodiversity and ecosystem researches. Detailed floristic analysis of benthic diatoms from the Crnica River has not been conducted before. The main aim of this work was to investigate the diversity and seasonal dynamics of epilithic diatom assemblages. Studies were conducted during 2011-2012 on the Crnica River, a right-side tributary of the Velika Morava River. Diatom frustules were cleaned using concentrated sulfuric acid (H<sub>2</sub>SO<sub>4</sub>), potassium permanganate (KMnO<sub>4</sub>) and oxalic acid. Permanent slides were made by Naphrax mounting medium, while the relative taxa abundance was determined by counting 400 valves on each slide. The diatom flora identified in the Crnica River was taxonomically diverse and included a total of 170 taxa representing 49 genera. The most frequent taxa found in almost all samples were: *Achnantheidium minutissimum*, *A. pyrenaicum*, *Amphora pediculus*, *Cocconeis placentula* var. *lineata*, *Gomphonema elengatissimum*, *Meridion circulare* and *Reimeria sinuata*. 24 taxa were defined as dominant. Their percentage contribution was 5% or more at least at one investigated site. Many of them were the most frequent taxa, also. During the investigated period, the highest number of taxa was recorded in the spring (139), and the lowest in autumn (94). Canonical correspondence analysis showed that a large number of taxa was only characteristic for the spring or winter period.

**KEYWORDS:** diatoms; biodiversity; floristic analysis; seasonal dynamics

Poster presentation 32 10 22

### DIVERSITY OF DIATOM COMMUNITIES IN THE VELIKA RUSANDA SODA PAN

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Species richness and diversity of diatoms inhabiting different niches (water column, sediment surface and submerged plants) as well ionic composition of water (Na<sup>+</sup>, K<sup>+</sup>, Mg<sup>2+</sup>, Ca<sup>2+</sup>, Cl<sup>-</sup>, SO<sub>4</sub><sup>2-</sup>, CO<sub>3</sub><sup>2-</sup>, HCO<sub>3</sub><sup>-</sup>) were studied in the Velika Rusanda soda pan during two years (late spring 2017 and 2018). These analyses are part of the preliminary study of diatom communities in soda pans in Serbia. The analysis of ionic composition confirmed previous finding that water of Velika Rusanda belongs to Na-HCO<sub>3</sub>-SO<sub>4</sub>-Cl chemical type. In 2017, a total of 15 taxa were identified, 5 in plankton and 14 in epiphytic community. In plankton the most abundant were *Craticula halophila* (36.25%) and *Anomoeoneis sphaerophora* (27.25%), while among epiphytic dominates *Surirella brebissonii* var. *kuetzingii* (46.25%). In 2018, a total of 22 taxa were identified, 3 in plankton, 19 in epiphytic community, and 14 on artificial substrate (brick). In plankton we recorded only a few individuals. The most dominant taxon in epiphytic community was *Nitzschia supralitorea* (95%), which also prevailed on brick (66.98%). The only genus recorded in 2017 was *Rhopalodia*, while genera presented only in 2018 were: *Cocconeis*, *Halamphora*, *Hantzschia*, *Humidophila* and *Planorhynchium*. Of the total number of identified taxa three were newly recorded for Serbian diatom flora: *Hantzschia weyprechtii* Grunow, *Navicymbulla pusilla* (Grunow) Krammer and *Nitzschia thermaloides* Hustedt. The most of identified taxa are characteristic for marine or brackish waters, as well as different types of freshwaters with elevated to highly elevated electrolyte content. The results of this preliminary study will help better understanding of diatom communities in soda pans and their assessment for conservation and restoration measures in these ecosystems.

**KEYWORDS:** shallow alkaline ponds, *Hantzschia weyprechtii*, *Navicymbulla pusilla*, *Nitzschia thermaloides*

Poster presentation 33 10 24

## DIATOMS ON THE CARAPACE OF EUROPEAN POND TURTLE (*EMYS ORBICULARIS* L.) IN VOJVODINA

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Epibiotic diatoms inhabiting the carapace of European pond turtle (*Emys orbicularis* Linnaeus, 1758) are examined between May and July 2017 from three locations in Vojvodina (Serbia): Special nature reserve „Deliblatska Peščara“, Natural monument „Ivankovačka ada“ and Special nature reserve „Gornje podunavlje“. Our objective was to compare the diatom flora on a single turtle species across part of its range. Diatoms were removed by brushing from the carapaces, preserved in 4% formalin and identified under Carl Zeiss AxioImager.M1 microscope after preparing permanent slides with Naphrax medium. The diverse diatom assemblages were recorded. A total of 116 diatom taxa from 32 genera were found on the carapace of European pond turtle. The highest diversity in epibiotic diatom community was recorded at SNP „Deliblatska Peščara“ and NM „Ivankovačka ada“ (85 and 81 taxa, respectively). Only 6 diatom taxa (*Nitzschia denticula*, *N. fonticola*, *Fragilaria ulna*, *Cymbella parva*, *C. perparva* and *Cyclotella* sp.) were found on all three localities. Of these species, *Cymbella perparva* i *Nitzschia denticula* were occurred on the greatest number of turtles. In contrast, 67 diatom taxa occurred only on a single locality. Diatom richness and abundance were variable both among and within localities. The most numerous were erect (*Cymbella* spp., *Gomphonema* spp.) and motile (*Nitzschia* spp., *Navicula* spp.) forms.

**KEYWORDS:** diatoms, epibionts, Deliblatska peščara, Ivankovačka ada, Gornje podunavlje

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