Presentation type: Oral Presentation, <u>Poster Presentation</u> (<u>underline the preferred type</u>) AMBRA1 controls plant development and senescence in *Physcomitrella patens*.

<u>Alessandro Alboresi¹</u>, Jessica Ceccato¹, Tomas Morosinotto¹, Luisa Dalla Valle¹. The first one should be the <u>presenting/corresponding author (underlined)</u>

¹Dipartimento di Biologia, Università di Padova, Via Ugo Bassi 58/B, 35121, Padova (<u>alessandro.alboresi@unipd.it</u>; <u>tomas.morosinotto@unipd.it</u>; luisa.dallavalle@unipd.it)

Autophagy is a universal mechanism that in plants control development, resistance to stresses and starvation. The role of autophagy is possible thanks to the programmed degradation of cell material that is delivered to the vacuole where hydrolases and proteases are localized. So far, many autophagy-related proteins (ATGs) have been identified. Some of them are universal, some are either specific to animals, plants or yeast. ATG protein complexes govern autophagosome initiation, nucleation, expansion, and maturation. In particular, the regulation of nucleation by the ATG6 (Beclin-1 in mammals) complex has not been well defined in plants.

Here we described the study of the <u>A</u>ctivating <u>M</u>olecule in <u>B</u>eclin 1-<u>R</u>egulated <u>A</u>utophagy (AMBRA1) protein, recently identified in mice and then characterized in our department in zebrafish and in the non-vertebrate chordate *Botryllus schlosseri*. In animals AMBRA1 is a positive regulator of autophagy that binds Beclin-1 upon autophagic stimuli. AMBRA1 is a large intrinsically disordered protein, able to bind other regulatory partners involved in cell processes such as autophagy, apoptosis, cell proliferation, development and cancer.

AMBRA1 sequence was found in plant genomes and we are studying its function in *Physcomitrella patens* where two lowly expressed genes are present, *AMBRA1a* and *AMBRA1b*. We have isolated single *ambra1a*, *ambra1b* and double *ambra1a/b* knock-out (KO) plants. The protonema of *ambra1a/b* KO showed growth defects as compared to WT, while *ambra1a* KO and *ambra1b* KO showed either no impairment or a slightly reduced growth respectively. *ambra1a/b* KO is characterized by a high frequency of aberrant branching pattern and a high percentage of senescent protonema cells both on minimum medium or on medium supplemented with glucose and ammonium tartrate. Moreover *ambra1a/b* KO showed unexpected vertical growth when cultivated in the light while it is unable of gravitropic growth when left in the dark for two weeks. Plants treated by protease inhibitors such as E-64d showed that the accumulation of degradation intermediates is not reduced in the absence of AMBRA1, meaning that potentially autophagy is still possible. AMBRA1 appears to be an important regulator of plant development, further studies will help to better understand the molecular function of AMBRA1 in plants.

Keywords: autophagy, Ambra1, tropism, ATG6, Beclin-1

Plitvice Lakes – the largest area with tufa forming bryophyte vegetation in Europe

Antun Alegro¹, Vedran Šegota¹, Anja Rimac¹, Nikola Koletić¹, Nina Vuković¹, Beáta Papp²

¹ Division of Botany, Department of Biology, Faculty of Science, University of Zagreb, Marulićev trg 20/II, 1000 Zagreb, Croatia (antun.alegro@biol.pmf.hr); ² Hungarian Natural History Museum, Könyves Kálmán krt. 40, 1087 Budapest, Hungary

Plitvice Lakes are situated in Dinaric region of Croatia characterized by different karst formations. One of them is the system of 16 larger and several smaller cascading lakes, stretched 8 km in length and interconnected by numerous waterfalls of different size, height and water velocity. They are formed through the biodynamic process of growing tufa barriers in water oversaturated with calcium carbonate, pH value higher than 8 and low concentration of dissolved organic matter. All these tufa barriers are overgrown by rich vegetation dominated by bryophytes. This vegetation was studied using phytosociological methods followed by measurements of environmental parameters and analysis of ecological indicator values. In total 44 bryophyte species were recorded of which 26 are constantly present in 90 vegetation relevés. The main characteristics of vegetation are prominent dominance of bryophytes over vascular plants and very constant species composition with a low number of species present with high abundances. Four main species are Palustriella commutata, Eucladium verticillatum, Hymenostylium recurvirostrum and Pellia endiviifolia. Nevertheless, three main types of bryophyte vegetation are differentiated – species poor vegetation of non-shaded waterfalls mostly with high water velocity, the vegetation of shaded waterfalls with weaker water flow, and species rich vegetation of moist rocks outside direct or constant influence of running water. All of these vegetation types belong to the moss rich vegetation of carbonate springs of montane and subalpine belts of Europe belonging to the alliance Cratoneurion commutati. However, due to the complex system of lakes and waterfalls, very large surfaces covered by this vegetation, species richness and their high abundances, Plitvice Lakes represent one of the most important diversity and distribution centres in Europe regarding the tufa forming bryophyte vegetation and is of the highest conservation value.

Keywords: Southeastern Europe, Cratoneurion, waterfall vegetation, bryophyte diversity

Antifungal effects of some mosses of Iran on the phytopathogenic fungus "Fusarium solani"

Roghaieh Asadboland¹, AliReza Iranbakhash¹, MohammadReza Esalahi², Saeed Shirzadian³

¹ Department of Biology, Science and Research Branch, Islamic Azad University, Tehran, Iran. (<u>roghaieh.asadboland</u>@srbiau.ac.ir). ² Plant Protection Department, Khuzestan Agriculture and Natural Resource Research and Education Center, Areeo, Ahvaz, Iran (mr_Eslahi@yahoo.com). ³ Department of Botany, Iranian Research Institute of Plant Protection, Agricultural Research Education and Extension Organization (AREEO), Tehran, Iran. (shirzadian2003@yahoo.co.uk)

Mosses have proved to have anti-cancer, anti-microbial and anti-fungal properties due to their specific chemical compounds. In this study, the antifungal effects of methanolic, ethanolic, acetonic and aqueous extracts from some mosses collected from different regions of Khuzestan province, Iran, were tested on growth of Fusarium solani, one of the most important pathogens of agricultural crops, especially wheat and compared to the commercial fungicide Benomyl. For this purpose, moss samples were transferred to the lab and washed with sterile water and dried in shadow. Then the dried mosses were pulverized and extracted by ethanol, methanol, acetone and distilled water. The antifungal activity was tested by disc diffusion method. Plates of Potato Dextrose Agar (PDA) were inculated with 6 mm diameter pieces of 7 days old fungal culture margin. Fungal pieces were placed in the middle of PDA medium. The filter paper discs (6 diameter) impregnated with different extracts and Benomyl were placed at a certain distance from the edge of fungus grown and kept in the incubator for 48 hours at 25 ° C. The diameter of the inhibition zone was measured using a millimeter ruler. In order to investigate the effect of moss extracts in vivo, wheat seeds of Chamran cultivar were implanted into moss extract and planted in pots containing 1: 10 mixture of soil and soil contaminated with fungi. Considering that the fungus species F. solani causes crown and root rot of wheat, after 35 days the root and crown of wheat plants were examined based on Wallwork scale (Wallwork et al. 2004). The experiment was conducted as a completely randomized design with three replications. The results show that ethanolic and methanolic extracts produced similar inhibitory effects as Benomyl fungicide, and aqueous extracts had limited effects. Statistical analysis using SAS 9.2 showed the significant variances between the effects of ethanolic extract and other extracts. In vivo observations also indicate that ethanolic extracts can prevent the growth of *F. solani* and in some cases, have similar effects as Benomyl.

Keywords: mosses, phytopathogenic fungi, antifungal effects, extract.

The bryophyte research programme at the Royal Botanic Garden Edinburgh (RBGE)

<u>Neil E. Bell¹</u>, Laura L. Forrest¹, Elizabeth Kungu¹

¹ Royal Botanic Garden Edinburgh, 20A Inverleith Row, Edinburgh, EH3 5LR, Scotland, U.K. (NBell@rbge.org.uk).

Bryophyte research at RBGE focuses on a range of activities integrating international taxonomic and phylogenetic research with the work of national conservation agencies and the long-established recording programmes of specialist societies to better understand the origins, diversity, distribution and future needs of the globally important Scottish bryophyte flora. British bryophytes are the best recorded in the world, while Scotland harbours much of Europe's remaining bryophyte-rich temperate rainforest and oceanic montane heath, host to many species with highly restricted and disjunct global distributions. RBGE further houses one of Europe's largest bryophyte herbaria (E) with over 500,000 specimens, including much recent, well-curated material and around 4,000 types.

Activities span core phylogenetic research on select groups (Polytrichaceae, thalloid liverworts, austral pleurocarps), large-scale molecular sequencing initiatives targeted at the British flora, and a number of smaller, self-contained projects addressing species-level problems in collaboration with international specialists and UK field bryologists. Our outreach work includes teaching bryophyte identification, contributing to "bioblitz" events, collaborating with visual artists and hosting regular visitors including secondary school and undergraduate students, while we are actively developing our collection of living bryophytes alongside our Horticulture Division.

Our ongoing liverwort DNA barcoding project includes multiple accessions of most British and Irish liverworts, providing a reference library for identification. Tangential to this are projects focusing on select taxa including *Aneura*, *Moerckia* and *Scapania*. Recent phylogenetic research highlights include an investigation of relationships between British species of the enigmatic, primarily austral genus *Orthodontium*, in which the native *O. gracile* is shown to belong to a distinct Holarctic and Asian clade that diverged from the rest of the genus early in the Cenozoic. This species has its last European outpost in Britain and appears to be threatened by the introduced *O. lineare*.

A major new project examining the global relationships of Scotland's oceanic liverwort heath community is now underway, aiming to explain the startling disjunctions displayed by many of the species by studying trans-continental phylogenetic structure, functional traits and dispersal capabilities.

Ongoing PhD projects include studies of *Polytrichum* sect. *Polytrichum* (Isuru Kariyawasam; University of Edinburgh), *Andreaea* and *Herbertus* (David Bell; University of British Columbia).

Keywords: taxonomy, DNA barcoding, Scottish liverwort heath, Orthodontium, herbarium.

Presentation type: Poster Presentation

Recommendations for the use of critical terms when applying IUCN red-listing criteria to bryophytes

<u>Ariel Bergamini¹</u>, Irene Bisang², Nick Hodgetts³, Neil Lockhart⁴, Jacques van Rooy^{5,6}, Tomas Hallingbäck⁷ [§]Co-first authors

¹ Swiss Federal Research Institute WSL, Zürcherstrasse 111, 8903 Birmensdorf, Switzerland (ariel.bergamini@wsl.ch); ² Swedish Museum of Natural History, Box 50007, SE-104 05 Stockholm, Sweden (Irene.Bisang@nrm.se); ³ Cuillin Views, 15 Earlish, Portree, Isle of Skye, IV51 9XL, UK (nick1901@hotmail.co.uk); ⁴ National Parks and Wildlife Service, Department of Culture, Heritage and the Gaeltacht, 90 North King Street, Dublin 7, D07N7CV, Ireland (<u>Neil.Lockhart@chg.gov.ie</u>); ⁵ National Herbarium, South African National Biodiversity Institute, Private Bag X101, Pretoria 0001, South Africa (<u>J.vanRooy@sanbi.org.za</u>); ⁶ School of Animal, Plant and Environmental Sciences, University of the Witwatersrand, Private Bag 3, Wits 2050, Johannesburg, South Africa; ⁷ Swedish University of Agricultural Sciences, SLU, Species Information Centre, Box 7007, SE-750 07 Uppsala, Sweden (<u>Tomas.Hallingback@slu.se</u>).

The IUCN Red List of threatened species (<u>http://www.iucnredlist.org/</u>) is widely recognised as an objective system to assess the extinction risk of animals, plants and fungi and as an authoritative tool to catalyse conservation actions. The IUCN criteria developed for estimating the extinction risk of species are most readily applied to large organisms with clearly identifiable sexually reproducing individuals, while they may be difficult to employ to bryophytes and other clonal and colonial organisms. Here, we present refined, pragmatic definitions for three key terms that are critical when using the IUCN red-listing methodology, notably 'mature individuals', 'generation length' and 'severe fragmentation'. These definitions facilitate the application of the IUCN Red List criteria for bryophytes. They have been successfully applied in the recent Red List assessment of all 1816 bryophyte species of Europe (Hodgetts et al., *in prep.*). The definitions provide an effective way of using the IUCN Red List criteria for bryophytes and may have a wider application to other clonal organisms.

Keywords: mature individual, individual-equivalent, severe fragmentation, generation length, IUCN Red List

Monitoring moss growth and physiological status using a low-cost image analysis system.

<u>Manuel M. Bustamante^{1,2}</u>, Indradatta deCastro-Arrazola², Belén Estébanez¹, Fernando Hurtado^{1,2}, Cristina Ronquillo², María Leo^{2,3}, Joaquín Hortal², Nagore G. Medina¹.

¹ Departamento de Biología (Botánica), Facultad de Ciencias, Universidad Autónoma de Madrid, Madrid 28049, Spain (manuel.molinab@uam.es); ² Department of Biogeography and Global Change, Museo Nacional de Ciencias Naturales (MNCN-CSIC), Madrid 28006, Spain; ³ Real Jardín Botánico (RJB-CSIC), Madrid 28014, Spain.

The last few decades there has been an increasing interest in using mosses as model organisms for ecological studies. This has brought a need for precise and easy-to-use methods to measure plant productivity and health. We present a non-destructive, cost- and time-efficient protocol for monitoring moss growth and physiological activity. The approach is based on the analysis of visible (VIS) and near infrared (NIR) moss pictures taken with low-cost drone cameras. Applying remote sensing and image analysis techniques we calculated a set of 19 spectral indices usually implemented in teledetection and 13 image segmenting methods. We used the ImageJ software and a modified version of the R package crustCover that we developed for these image analyses. Then we evaluate which index-segmentation combination provides higher precision and accuracy for moss area estimation. Finally, we estimate the moss physiological status using the segmented images and the most meaningful indices. To calibrate the protocol, we have used five pictures per moss species, and six common soil moss species (Dicranum scoparium, Hypnum cupressiforme, Homalothecium aureum, Ptychostomum capillare, Tortella squarrosa and Syntrichia ruralis), which were cultured for six months in 2018 in three field localities along an altitudinal gradient in central Spain. The most informative indices to estimate moss area and physiological status were NDVI, MSAVI, EVI and Saturation. We found differences in the efficiency between methods to estimate moss areas automatically, depending on the relationship between moss and background area in the pictures. This fact calls for a careful use of automatic segmentation, highlighting the importance of manually adjusting the index thresholds on our subject of study. The future implementation of neural networks and deep learning on methods based on spectral indices provides a wide perspective for automated data collection in the field or in the laboratory.

Keywords: Moss growth, image analysis, remote sensing, spectral indices.

Elucidating the role of eIF5A in growth and stress response in Physcomitrella patens

Alexandra Castro, Claudia Deluca, Cecilia Ruibal and Sabina Vidal

Laboratorio de Biología Molecular Vegetal, Facultad de Ciencias, Universidad de la República, Montevideo, Uruguay. (acastro@fcien.edu.uy)

The control of protein translation is a central aspect of adaptive responses to environmental stress in plants, as well as in development. Translation initiation is a highly regulated and rate-limiting step of protein synthesis that requires more than 12 eukaryotic initiation factors (eIFs).

In this study, we have used RNA-seq to identify genes involved in translational regulation of gene expression during high temperature stress in *Physcomitrella patens*. A gene enconding a eIF5A protein homolog (*PpeIF5A*) was shown to be highly induced in response to high temperature. PpeIF5A encodes a 17.5 kDa deduced protein, with a theoretical pl of 5.60 and a predicted signal peptide. In planta transient expression of PpeIF-GFP fusion protein indicated that PpeIF5A localized in the nucleus of plant cells under non stress conditions. Expression profiling of this gene showed that, in addition to heat stress, PpeIF5A was upregulated in response to dehydration, abscisic acid and salicylic acid treatments.

5A translation initiation factors have been suggested to play an important role in the selective translation of proteins that, due to their biochemical nature, are difficult to be translated. Among the possible target genes, whose expression are expected to depend on eIF5A, are those that encode proteins rich in PPP (Pro-Pro-Pro) and/or PPG (Pro-Pro-Gly) sequences. A search in *P. patens* genome revealed the presence of a number of putative target genes. The expression profile of those genes was analyzed in RNA-seq transcriptomic data, in response to heat and salicylic acid treatment. The aim of this work is to characterize the function of PpeIF5A and to identify the proteins involved in stress responses that require the activity of this factor for proper translation.

Keywords: eIF5A, hypusine, translation, polyproline

Population genetic diversity, genetic structure and phylogeography of *Riella helicophylla* (Riellaceae)

José Gabriel Segarra-Moragues^{1,2}, Manuel Jesús Gil-López^{1,3}, Felisa Puche^{1,4},

¹ Departamento de Botánica y Geología, Universidad de Valencia, Burjassot (Valencia), E-46100, Spain; ² (j.gabriel.segarra@uv.es); ³ (manuel.j.gil@uv.es); ⁴ (m.f.puche@uv.es).

Riella helicophylla (Bory & Mont.) Mont. (Riellaceae, Sphaerocarpales) is an aquatic, liverwort of brackish, temporary Mediterranean lagoons. It is a dioicous, ephemeral species, with highly fluctuant populations, which relies on spores for population regeneration after summer droughts. Nonetheless, it can also spread vegetatively during the growing season due to propaguliferous scales. It has a widespread fragmented distribution in Mediterranean countries and mainland Spain is where the largest number of populations have been reported. The species is considered threatened and included in Red Lists of the corresponding countries and in the Annex II of the European Union Directive. The species is being studied both morphologically and molecularly as earlier studies have shown it consists of at least two taxa. Here we present the results of a population genetic study aimed at describing the genetic diversity and population structure within and among populations and phylogeographical patterns of this liverwort. For this purpose 756 individuals from 26 populations of the most widespread taxon of the R. helicophylla complex, covering the whole Spanish range and of some of the western Mediterranean countries have been genotyped at 9 nuclear microsatellite loci specifically designed for the species. From this analysis, 90 alleles were detected showing moderate to high levels of genetic diversity (overall genetic diversity among individuals across populations ranging from 0.148 to 0.532). Distance based clustering and Bayesian analysis of population structure showed a strong genetic association of populations within areas. However, geographical areas with longer fluctuations on habitat suitability or more prone to shorter flooding times, showed higher among population genetic structure within geographical areas. This may be related to temporal variation of population development thereby affecting population connectivity through migratory routes of aquatic birds.

Acknowledgments: We acknowledge financial support by the Ministry of Economy industry and Competitiveness (project CGL2016-80418-P).

Keywords: Aquatic liverworts, endangered species, phylogeography, Riellaceae

Mosses of Central Florida: Current progress in their documentation and in development of new identification tools

Frederick B. Essig¹, Ainun Nadhifah^{1,2}

¹USF Herbarium, University of South Florida, Tampa, FL 33620, U.S.A. (<u>essig@usf.edu</u>); ²Cibodas Botanic Gardens, Indonesian Institute of Sciences (LIPI), PO BOX 19 Sindanglaya, Cipanas, Cianjur, Jawa Barat 43253, Indonesia.

Central Florida is a region with a distinctive floristic composition, where the mosses from temperate North America and the neotropics are interfused. Presently we review the recent status of the documentation of the mosses of Florida and progress in the development of modern interactive identification tools for Central Florida. The primary database is in the Atlas of Florida Plants, based on the species listed in the Flora of North America (FNA) as occurring in Florida, with county distributions of moss species collected from the database of the Consortium of North America Bryophyte Herbaria (CNABH). Development of new identification tools include species portraits on the Botany professor blogsite and a new interactive key to the mosses of Central Florida. Data reveal that Seminole County is the most heavily collected county with 132 species out of 264 listed in the FNA. Analysis of species abundance shows the three most common species found in Central Florida are *lsopterygium tenerum, Leucobryum albidum*, and *Octoblepharum albidum*. To date, 55 mosses have been incorporated into the key and 53 moss portraits have been posted. This project is still in progress. The publication of innovative identification tools is intended to increase curiosity and encourage others to study the mosses of this region.

Keywords: Mosses, identification, interactive key, Central Florida, bryophytes.

The effect of lead pollution on chlorophyll fluorescence in two moss species

<u>Belén Estébanez¹</u>, Helena Palancar¹, Patricia Haro²

¹ Departamento de Biología, Facultad de Ciencias, Universidad Autónoma de Madrid, Madrid 28049, Spain (belen.estebanez@uam.es, helena.palancar@estudiante.uam.es); ² Departamento de Física de Materiales, Facultad de Ciencias, Universidad Autónoma de Madrid, Madrid 28049, Spain (patricia.haro@uam.es).

Heavy metals may produce structural and reproductive alterations in mosses. Their biomonitoring potential depends on the relationship of the extent of these effects and the pollutant level in the environment. In order to develop a non-invasive, easy measurement of these alterations on mosses, we present here an analysis of chlorophyll fluorescence, to test the effect of experimentally supplied lead on two moss species, Homalothecium aureum and Tortula muralis, with contrasted growth forms and ecological requirements. We applied three doses of $Pb(NO_3)_2$ and a control with water on greenhouse cultures of each species (6 replicae per experiment). The doses were sprayed twice every week during two months. We analysed chlorophyll fluorescence using two techniques: 1) fluorescence imaging using a CCD camera, and 2) spectroscopic analysis of the emission spectrum. The chlorophyll fluorescence was excited by using a low-power laser beam (405 nm, 10 mW), and detected in the red region of the spectrum (600-700 nm) by placing several filters. Both techniques provide us information about the chlorophyll content, which is affected by the lead pollution. Initially *H. aureum* presents a higher chlorophyll content, and apparently adjusts better to culture conditions before lead spraying. After the lead supply experiment, the specimens of both species survive, although visually H. aureum showed more conspicuous damages (yellowing), whereas T. muralis presented little apparent alterations, and was able to produce new sporophytes. The fluorescence results point at a reduction of fluorescence in *H. aureum* in higher lead doses, more remarkable than in T. muralis. We interpret that T. muralis is more naturally resistant, and that H. aureum shows a greater extent of adaptation of its photosystems to pollution stress. These techniques allows a non-invasive, periodical monitoring of the effects of pollutants on mosses, showing pollutant effects on chlorophyll and possible coping mechanisms of pollution-stressed plants.

Keywords: chlorophyll fluorescence, lead pollution, outdoor cultures

Dust in the wind... How to improve air quality monitoring using terrestrial mosses.

<u>R. García-Seoane¹</u>, J.A. Fernández¹, J.R. Aboal¹

¹Ecology Unit, Dept. Functional Biology, Universidade de Santiago de Compostela, Fac. Biología, Lope Gómez de Marzoa s/n, Santiago de Compostela, 15702, A Coruña, Spain.

Terrestrial mosses have been used for more than 40 years to monitor trace elements in atmospheric deposition because of their high cation exchange capacity and suitable morphology for fixing and accumulating particulate and gaseous pollutants. The most common type of active biomonitoring using moss transplants is the "moss bag technique", which consists of the exposure of moss samples held within mesh bags and the subsequent analysis of contaminant concentrations in their tissues. Standardized protocols have unfortunately still not been developed and research has mainly focused on evaluating methodological variables such as the selection of species and the shape and size of the moss bag. However, it has been suggested that the orientation of the bags relative to a pollution source may greatly influence the efficiency of interception and capture of atmospheric contaminants by moss. Thus, the objective of this study was to investigate whether the optimal orientation of flat moss bags containing the moss Sphagnum palustre improves the retention of Al, As, Cd, Cr, Cu, Fe, Hg, Ni, Pb, V and Zn. Three replicate moss bags were exposed in each of eight industrial sites (in the surroundings of two ferrous-smelters, a ceramics factory, a paper and wood factory with cogeneration production, and a coal-fired power plant) in two different ways: attached to weathervanes (bags in devices rotating downwind) and attached to static poles (static bags). The results demonstrate that in the most polluted sites, the metal uptake capacity (for As, Cu, Fe, Pb and V) in the moss bags attached to the weathervanes was higher than in the moss bags attached to the static poles, so the moss bags attached to the weathervanes were more effective for detecting pollution than the others. Data suggest that the orientation of the bags with respect to the source of contamination should be considered and that further research is required to standardize the methods used in the moss bag technique.

Keywords: atmospheric pollution, biomonitoring, heavy metals, mosses

The importance of ash stands for the diversity and conservation of epiphytes in broad-leaved forests of Latvia.

Linda Gerra-Inohosa¹, Līga Strazdiņa², Ilze Matisone¹

¹Latvian State Forest Research Institute "Silava"; Rigas Street 111, Salaspils, LV-2169, Latvia (linda.gerra@silava.lv); ² University of Latvia, Botanical Garden, Kandavas Street 2, Riga, LV-1083, Latvia (liga.strazdina@lu.lv).

In recent years, a rapid destruction of the European ash Fraxinus excelsior caused by the ascomycete Hymenoscyphus fraxineus has been observed throughout Europe. Latvia, located in northern part of Europe in the boreo-nemoral vegetation zone, has been exposed to ash dieback as well, which has resulted in decreasing habitats for many epiphytic bryophyte species. The objective of this study was to determine the diversity of epiphytes in broad-leaved forests, highlighting the flora of bryophytes in *Fraxinus excelsior* forests. The data were collected during previous four summer seasons (from 2015 till 2018). Epiphytic bryophytes were examined in 62 forest stands throughout the territory of Latvia. The stands were classified into five groups according to dominant tree species. The results of this study indicate that ash stands were hosting the highest species richness. Overall, 82 bryophyte species were recorded on 848 tree trunks, including 14 especially protected species for Latvia. Among these, 70 species were found in ash stands, of which 13 epiphytes were recorded only in plots of *Fraxinus excelsior* forests. The number of species per plot were significantly higher in ash stands (24 species/plot) compared to lime (15), oak (14), hornbeam (14) and elm (10) stands. Ash stands were characterized also by the highest abundance of protected species (in total, 12 species). The composition of epiphytes does not differ significantly between different forest stands. However, 23 epiphyte species showed their preference to ash forests, including two protected species – Homalia trichomanoides, Anomodon longifolius and one rare species in Latvia – Frullania fragilifolia. According to the results, the lime and oak stands had the highest proportion of epiphyte species that were also recorded in ash stands. In conclusion, it could be assumed that ash stands in Latvia represent high conservation value for epiphytes. The recommendations for forestry politics to preserve old oak stands in Latvia might compensate decreasing ash substrate in future perspective.

Keywords: epiphyte diversity, broad-leaved forests, ash stands, species richness

Information for submission of abstracts to the Bryology 2019 meeting. Deadline: 31 May 2019

Presentation type: Oral Presentation, Poster Presentation (underline the preferred type)

Abstract Title: Spatial analysis of qualitative pollution data in moss samples at a regional scale.

<u>Giráldez P</u>.¹, Varela Z.², Crujeiras R.M.¹, Aboal J.R.¹, Fernández J.A.¹

¹ Grupo de Ecotoxicología, Departamento de Biología Funcional, Facultad de Biología, Universidad de Santiago de Compostela, 15782 Santiago de Compostela, Spain (P.G.: <u>pablogiraldez4b@gmail.com</u>, <u>Z.V.: zulema.varela@usc.es</u>, J.R.A.: <u>jesusramon.aboal@usc.es</u>, J.A.F: <u>jangel.fernandez@usc.es</u>);
³ Departamento de Estadística, Análisis Matemático y Optimización, Facultad de Matemáticas, Universidad de Santiago de Compostela, 15705 Santiago de Compostela, Spain (<u>rosa.crujeiras@usc.es</u>)

Terrestrial mosses have been used as air pollution biomonitors for more than 40 years, but despite this, and due to certain technical limitations, moss biomonitoring has hardly had any impact or has influenced decision-making regarding environmental pollution. One of the causes of this is the scarce bibliography or theoretical knowledge about the process of accumulation of pollutants by the mosses and the real relationship between atmospheric deposition and the concentrations of pollutants found in moss tissues, which makes it difficult to interpret the results. Even so, most of the studies carried out in this field carry out a quantitative analysis of the concentrations, which clearly seems to be contraindicated. The objective of the present study is the analysis of the spatial structure of various pollutants in Galicia (NW Spain) through a new statistical procedure. This procedure is presented as a new possibility for the qualitative treatment of quantitative data on the concentration of pollutants in terrestrial mosses. This analysis proposes a new allocation rule for the categorisation of quantitative data into the dichotomous variable of "polluted" and "uncontaminated" levels. This rule is based on the hypothesis that the concentration of heavy metals in the moss, in the absence of pollution, follows, like the nutrients, a normal distribution. In this study we categorized moss samples collected in the 150 points of Galicia sampled in spring and autumn of different years. Once the data had been categorised, the spatial analysis was carried out, calculating the indicator semivariograms and obtaining the predictions by means of indicator krigging. The results showed the existence of dependence and spatial structure for some metals such as Hg, where significant differences in pollution can be observed between different points of the study area, which is in line with the results observed in the bibliography. Based on this, we can conclude that this seems to be a suitable procedure for the qualitative treatment of the data, although this procedure could not in itself determine whether a point is contaminated or not, but rather determine whether it is contaminated with respect to the background contamination level.

Keywords: air pollution, bryophytes, moss bag technique, spatial analysis.

Bryology 2019, Madrid, Spain, July 9-12, 2019

Presentation type: Poster Presentation

Bryophyte Phylogeny Poster (BPP)

Theodor C. H. Cole^{1*°}, Hartmut H. Hilger^{2*°}, <u>Bernard Goffinet</u>^{3*}, Hiroyuki Akiyama⁴, Oyuntsetseg Batlai⁵, Sahut Chantanaorrapint⁶, Denise Pinheiro da Costa⁷, Nyambayar Dashzeveg⁸, Enkhjargal Enkhtaivan⁹, Johannes Enroth¹⁰, Zigmantas Gudžinskas¹¹, Ida Haerida¹², Xiaolan He¹³, Lars Hedenäs¹⁴, Michael Ignatov¹⁵, Rafael Medina¹⁶, Svitlana O. Nyporko¹⁷, Ryszard Ochyra¹⁸, Elizabeth A. Widjaja¹⁹, Wen Ye²⁰, Li Zhang²¹

^{1,2} Dahlem Centre of Plant Sciences (DCPS), Institute of Biology – Botany, Freie Universität Berlin, Altensteinstr. 6, D-14195 Berlin, Germany (t.c.h.cole@fu-berlin.de; Hartmut.Hilger@fu-berlin.de); ³ Department of Ecology and Evolutionary Biology, University of Connecticut, 75 North Eagleville Rd, Storrs CT, 06269-3043, U.S.A. (bernard.goffinet@uconn.edu); ⁴ Museum of Nature and Human Activities, Hyogo 6 chome, Yayoigaoka, Sanda, Hyogo 669-1546, Japan (akiyama@hitohaku.jp); ^{5,8} Department of Biology, School of Arts and Sciences, National University of Mongolia (oyunaa@num.edu.mn; nyambayar@num.edu.mn); ⁶ Dept. of Biology, Prince of Songkla University, 15 Karnjanavanich Rd. Hat Yai, Songkla 90110, Thailand (sahut.c@psu.ac.th); 7 Jardim Botânico do Rio de Janeiro, Rua Pacheco Leão 915, 22460-030, Rio de Janeiro, RJ, Brasil (dcosta@jbrj.gov.br); ⁹ Laboratory of the Flora and Plant Systematics, Institute of General and Experimental Biology, Mongolian Academy of Sciences, Peace Ave., Bayanzurkh District, Ulaanbaatar 13330-0005, Mongolia (eegii88@gmail.com); ¹⁰ Organismal and Evolutionary Biology Research Programme, Faculty of Biological and Environmental Sciences, P.O. Box 65, FI-00014 University of Helsinki, Finland (johannes.enroth@helsinki.fi); ¹¹ Institute of Botany of the Nature Research Centre, Žaliųjų Ežerų Str. 49, LT-08406 Vilnius, Lithuania (zigmantas.gudzinskas@gmail.com); 12,19 Herbarium Bogoriense, Botany Division, Research Center for Biology, LIPI, Cibinong Science Center, Jl. Raya, Jakarta-Bogor Km. 46, Cibinong, 16911, Bogor, Indonesia (ihaerida@gmail.com; eawidjaja3003@gmail.com);¹³ Botany Unit, Finnish Museum of Natural History, P.O. Box 7, FI-00014 University of Helsinki, Finland (xiao-lan.he@helsinki.fi);¹⁴ Dept. of Botany, Swedish Museum of Natural History, Box 50007, 10405 Stockholm, Sweden (lars.hedenas@nrm.se); ¹⁵ Lomonosov Moscow State University, Biological Faculty, Geobotany Dept., Leninskie Gory Str. 1-12, Moscow 119234, Russia (misha_ignatov@list.ru); ¹⁶ Augustana College, Department of Biology, 639 38th Street, Rock Island, IL 61201, U.S.A. (rafaelmedina@augustana.edu); ¹⁷ M.G. Kholodny Institute of Botany, National Academy of Sciences of Ukraine, 2 Tereshchenkivska Street, Kyiv (Kiev) 01004, Ukraine (nyporkos@ukr.net);¹⁸W. Szafer Institute of Botany, Dept. of Bryology Polish Academy of Sciences, ul. Lubicz 46 31-512, Kraków Poland (R.Ochyra@botany.pl); ²⁰ South China Botanical Garden, Chinese Academy of Sciences, 723 Xingke Road, Tianhe District, 510650 Guangzhou, China (wenyescib@scbg.ac.cn); ²¹ Fairy Lake Botanical Garden, Shenzhen & Chinese Academy of Science, 160 Xianhu Road Liantang, 518004 Shenzhen, Guangdong, China (zhangli@scib.ac.cn); * corresponding authors, contributed equally; ° conceptually designed and created the poster

The educational **Bryophyte Phylogeny Poster** (BPP) provides a graphic overview of the relationships among Bryophytes s.l. (liverworts, mosses, hornworts) with divisions, major subclasses, orders, and families along with relevant apomorphic and plesiomorphic traits. This depiction of our current understanding of phylogenetic relationships is an ideal tool designed for teaching, studying, and research.

The original poster is available for free downloading from PeerJ Preprints at https://peerj.com/preprints/27571/ in its most recent updated version. The BPP sofar has been translated into 16 languages (English, German, Spanish, Portuguese, French, Chinese, Japanese, Mongolian, Thai, Indonesian, Russian, Ukrainian, Polish, Swedish, Finnish, and Lithuanian). All versions are freely available to the international community via ResearchGate and via the FU Berlin online webpages. Regular updates of the posters will be made as new research and data are published.

Keywords: Bryophytes, phylogeny, poster, education, teaching

Towards a database of the Romanian bryophyte vegetation. Status and perspectives.

<u>Goia I.¹*,</u> Şuteu A.¹

¹ Babeș-Bolyai Univ. of Cluj-Napoca, Republicii Str. 44, Cluj-Napoca, Romania, RO-400006.

A vegetation database represents a valuable resource for the continuous adjustment of the syntaxonomical system, as well as inestimable information about species coenology, synecology and syndynamics, particularly for species of conservative interest. Our aim is: 1. to compile all phytosociological data from Romania into a database which will be related to other existing inventories, e.g. Romanian Grassland Database (RGD), integrated into the Global Index of Vegetation-Plot Databases (GIVD); 2. to review the coenotaxonomic knowledge on bryophyte communities according to the new European syntaxonomical framework (Mucina et al., 2016); 3. to identify the bryophyte communities hosting bryophytes of conservative interest.

The first step was to assemble all epiphytic relevés from the literature and to add them to our own data. The nomenclature follows Grolle & Long (2000) for liverworts and Hill et al. (2006) for mosses. Syntaxonomical classification is a minute and complex process: in a first phase, updates (added as new columns) were made according to Marstaller, 1992.

The epiphytic database comprises 620 relevés: 574 are from the literature (16 papers and two PhD theses) and 46 relevés are unpublished. Surveys were conducted on 31 tree species (with a diameter between 15-180 cm); most of the relevés were sampled on beech, pedunculate and sessile oak, and only 4.03% were sampled on coniferous bark. Concerning the vegetation classes, corticolous moss communities are best represented by *Neckeretea complanatae* Marst. 1986 and *Frullanio - Leucodontetea sciuroidis* Mohan 1978 em. Marst. 1985, and on acid or old bark, by *Cladonio - Lepidozietea reptantis* Jez. et Vondr. 1962 em. Marst. 1993. Ten species are of conservative interest: *Dicranum viride* (HD/BC/EN), *Lescuraea mutabilis, Metzgeria violacea, Neckera pennata, N. pumila* and *Zygodon viridissimus* are vulnerable, *Habdrodon perpusillus, Hypnum jutlandicum, Leptodon smithii* and *Pseudoleskea saviana* are critically endangered.

Data gaps are inherent to such a large study (e.g. elevation, aspect, host species, sample area, tree diameter, GPS coordinates or location). Some relevés were not assigned to any association. We plan to analyze and improve the syntaxonomical knowledge about epiphyte communities from Romania and then to extend the available data about bryophyte communities.

Keywords: Epiphyte communities, database, red listed species.

Competition and facilitation in the early development of three common Iberian mosses

Miguel Ángel Gómez-Molinero, Nagore G. Medina and Belén Estébanez

Departamento de Biología (Botánica), Facultad de Ciencias, Universidad Autónoma de Madrid, Campus de Cantoblanco, C/Darwin 2, Madrid, Spain

(miguelangel.gomezm@estudiante.uam.es; nagore.garcia@uam.es; belen.estebanez@uam.es)

Although biotic interactions affecting the spore germination may play a major role in the establishment of a moss community, they are rarely documented and their extent is not well known. In order to analyze the effects of shoots and spores of mosses on germination and early protonematal development, we have modified the sandwich technique, a widely used in vitro assay that was designed to assess allelopathic effects in vascular plants, and adjusted it for living bryophyte material. We have performed intra- and interspecific assays with three common mosses in central Spain: Hypnum cupressiforme Hedw., Tortula muralis Hedw. and Dicranum scoparium Hedw., using both spores and disinfected vegetative apical shoots to test their effects on spore germination and growth. We have obtained different patterns of response for each species. The protonemata of *H. cupressiforme* are inhibited only by its own spores and by shoots of D. scoparium (both co-habiting in their natural environment), but not by T. muralis (a non-coexistent species). In turn, T. muralis is inhibited by both the spores and shoots of *H. cupressiforme*, and by the presence of shoots of *D. scoparium*. Strikingly, the sporeling development of D. scoparium, although quite limited in control experiments, is enhanced, not inhibited, by the presence of spores or shoots of any of the three species (most especially, by shoots of *H. cupressiforme*). We have found that the effect of shoot leachate is similar to shoots themselves, suggesting that the facilitation or inhibitory effects may depend either on a stable, water-carried factor, or on the development of shoot epiphytic communities of micro-organisms that interact with spore germination and growth. Our results in these three species point at several interesting implications: 1) a pre-colonization requirement for the spores of *D. scoparium*, combined with an inhibitory strategy of their shoots over the establishment of new mosses; 2) the occurrence of spore-spore intraspecific competition in *H. cupressiforme*, but not between adults and spores; and 3) the sensitivity of Tortula muralis even to naturally non-coexistent species. These interactions, usually neglected in bryophytes, may play a crucial role in their distribution and dynamics.

Keywords: Bryophyte interactions, in vitro culture, facilitation, competition, spore germination.

Con formato: Sin subrayado Con formato: Subrayado

Sporophyte and spore characters in three <u>genus ogenera o</u>f difficult morphological characterisation of <u>within</u> the family Brachytheciaceae

Daniel Guerra-Mateo¹, Belén Estébanez², José David Orgaz³

¹ Universidad Internacional Menéndez Pelayo, Madrid, 28040 Spain (guerra.mateo.daniel@gmail.es); ² Department of Biological Sciences, Universidad Autónoma de Madrid, Madrid, 28049, Spain (belen.estebanez@uam.es); ³ Department of Biological Sciences, Universidad de Murcia, Murcia, 30003, Spain (jdavid.orgaz@um.es).

The traditional genus Brachythecium, the most important taxonomic group in the family Brachytheciaceae, was split on molecular grounds into three monophyletic genera: Brachythecium s. str., Brachytheciastrum and Sciuro-hypnum. Morphological characters, usually more centred on the gametophyte, yield little support for this phylogenetic classification. However, the sporophyte and the spores of this group remain poorly addressed. Here we present an anatomical and ultrastructural study on the sporophyte and spores in this group in order to find some sound characters allowing morphological support for the molecular classification. We have studied either freshly collected samples or herbarium specimens of 20 Palaearctic species of Brachythecium s.l., and of 4 additional external species (two in Brachytheciaceae, and two in other families of O. Hypnales). We have observed them using both light and electron microscopy: scanning electron microscopy for spore ornamentation and peristomes, and transmission electron microscopy for the spore inner structure. Our results provide novel descriptions for the sporophyte and spore ultrastructure for the species studied. Besides, we could identify the most valuable sources of morphological characters supporting the molecular circumscription of the group, namely the anatomy of the seta and the sporoderm inner structure. In Brachytheciastrum the inner cortical cells in the seta are distinctively small, in Brachytheciastrum s. str. the cell walls of this layer are thinner, and in Sciuro-hypnum most of the internal seta structure corresponds to a thick-walled cortex. In turn, the spore in Brachytheciastrum has an isopolar, thin sporoderm; whereas in Brachythecium the intine is thicker, with marked polarity, and in Sciuro-hypnum it is even thicker and highly heteropolar.

Keywords: Brachytheciaceae, morphology, ultrastructure, seta anatomy, spore.

Green workshops - a focus on biodiversity using bryophytes for urban children

<u>Christine Habashi¹</u>, Romain Dewaele²

^{1,2} Conservatoire et Jardin botaniques de la Ville de Genève, Geneva, Switzerland (Christine.habashi@ville-ge.ch)

One of the missions of the Conservatory and Botanical Garden of Geneva (CJBG) is to transmit our knowledge on plant biodiversity, and the importance of biodiversity for our planet, to the public. As an institution that holds rich bryophyte collections (Hedwig-Schwägrichen, Stephani) we have been seeking ways to inform on biodiversity using bryophytes through our Green Workshops programme. The Green Workshops are open to children from Geneva from 9-12 years old and represent a rich collaboration between the scientists and education staff of the CJBG with members of the University of the Third Age (U3A) of Geneva. The aim of our newly developed workshop was to introduce children to bryophytes – an often integral part of the flora of cities – and to scientific enquiry in the field of biodiversity. We also wanted to demonstrate some of the science done on bryophytes at our institution. To bring bryophytes to life in a playful way for urban children, an activity based on the search for a 'lost' moss professor was created. The children needed to find Professor "Mousse-tach'" using her notebook on bryophytes and by finding clues or answering questions to fill in parts of the notebook that were blank. In groups, the children visited different 'exploration stations' that were run by the workshop team. Each station specialist demonstrated something about bryophytes (What is a bryophyte? What are their main features? Where do they grow? How do they survive?). The notebook itself, created in-house and distributed to each child, contains historical drawings, brief explanations about bryophytes and spaces for them to add pictures or specimens of bryophytes. When the children had found the lost professor, they had to search for some common species within the botanical garden grounds with her using their hand-lenses. To see bryophytes close-up and to get a taste for the scientific study of bryophytes, the children then went into the laboratory and observed bryophytes using stereoscopes working through a key to determine six different species. Once they had identified their material, they added it to the notebook to make a mini-herbarium that they could take home.

Keywords: Education, outreach, bryophytes, city flora

Investigating the Biosynthesis and Function of Complex Sphingolipids in Physcomitrella patens

Jasmin Gömann¹, Hanno Resemann¹, <u>Tegan M. Haslam</u>¹, Cornelia Herrfurth^{1,2}, Kirstin Feussner^{1,2}, Ellen Hornung¹, Ivo Feussner¹

¹Georg-August-University, Albrecht-von-Haller-Institute for Plant Sciences, Goettingen Center for Molecular Biosciences (GZMB), Dept. of Plant Biochemistry, D-37077 Goettingen, Germany; ²Georg-August-University, Goettingen Center for Molecular Biosciences (GZMB), Service Unit for Metabolomics and Lipidomics, D-37077 Goettingen, Germany; <u>tegan.haslam@biologie.uni-</u> <u>goettingen.de</u>

Sphingolipids are essential and characteristic membrane components of eukaryotic cells. In addition, sphingolipids are signalling molecules. They contribute to membrane integrity, are required for normal growth and development, and mediate plant-pathogen interactions. There are two broad classes of complex sphingolipids found in plant cells, glucosylceramides (GlcCers) and glycosyl inositolphosphorylceramides (GIPCs), which both consist of a hydrophobic ceramide backbone and a polar sugar head group. Both GlcCers and GIPCs have proven difficult to study due to the technical challenges of extracting amphipathic lipids, and genetic challenges posed by the fact that GlcCerand GIPC-deficient mutants are either lethal or have severely impaired development. The model bryophyte *Physcomitrella patens* is an appealing system for the study of sphingolipid metabolism, as mutant phenotypes may be easier to interpret within its relatively minimalist developmental program compared to the conventional angiosperm model Arabidopsis thaliana. Further, a model bryophyte presents an opportunity to study the most ancestral functions of complex sphingolipids in land plants. We have profiled the sphingolipid composition of Physcomitrella patens by ultraperformance liquid chromatography coupled to mass spectrometry (UPLC-MS/MS). Unlike other plant species in which GIPCs are highly abundant, Physcomitrella patens accumulates a high level of GlcCers, while GIPCs are barely detectable. We identified candidate genes associated with both GlcCer and GIPC biosynthesis in the *Physcomitrella patens* genome based on homology to Arabidopsis thaliana. We are confirming these annotations with biochemical assays, and by generating and characterizing loss-of-function mutants. We generated a ceramide glucosyl transferase (cgt) mutant by homologous recombination; this mutant lacks GlcCers entirely, and accumulates ceramides. The mutant has abnormal growth, and appears unable to produce caulonema in a dark-growth experiment. Single and higher-order inositol phosphorylceramide synthase (ipcs) mutants are being generated using CRISPR/Cas9. This work sheds light on the evolution of the metabolism of an enigmatic class of lipids, and contributes to our understanding of their cellular and developmental functions.

Keywords: Physcomitrella patens, lipid metabolism, sphingolipids, CRISPR/Cas9 mutagenesis

Of turf, trees and air quality: does roadside moss trap more particulate matter than leaves?

Alison Haynes^{1,2}, Robert Popek^{2,3}, Mitchell Boles^{1,4}, Clare Paton-Walsh^{1,4}, and Sharon A. Robinson

- ¹ School of Earth, Atmospheric and Life Sciences (SEALS), University of Wollongong, NSW 2522, Australia; amh304@uowmail.edu.au
- ² Centre for Sustainable Ecosystem Solutions, University of Wollongong, NSW 2522, Australia;
- ³ Laboratory of Basic Research in Horticulture, Faculty of Horticulture, Biotechnology and Landscape Architecture, Warsaw University of Life Sciences - SGGW, Warsaw 02-787, Poland;
- ⁴ Centre for Atmospheric Chemistry, University of Wollongong, NSW 2522, Australia;

Plants in urban areas interact with air quality in numerous ways. Firstly, pollutants from industry, vehicular and residential sources can be detrimental to plant growth. Secondly, plants sometimes contribute to poor air quality, for instance by emitting allergens such as pollen, or by trapping pollutants in street canyons and thirdly, with appropriate placement some species improve air quality through phytoremediation. A common urban pollutant is particulate matter (PM - small particles of solid or liquid). While this is of concern to human health, less well known is its effect on vegetation and while moss is commonly studied as a biomonitor, there is little research on how it is affected by urbanisation. Our objective was to measure PM entrapment by roadside moss turfs and compare it to leaves of a common Australian tree species, *Pittosporum undulatum* on an urban gradient. We also wished to compare stress levels on this gradient using chlorophyll fluorescence (F_v/F_m) . We sampled nine sites in the coastal city of Wollongong, NSW, three in each of three levels of urbanisation: low, medium, and high according to road type (freeway, suburban road, quiet periurban road). At one site of each urban class we measured $PM_{2.5}$ over a two-week period using a mobile monitor. PM of three size fractions was isolated by filtration and washing with both water and chloroform. Site averages for moss turfs were much higher than leaves: between 5.60 and 33.00 mg per g dry weight for total PM (moss) compared to between 2.15 and 10.24 mg per g dry weight (tree). Moss appears to be more sensitive to increasing urbanisation in terms of photosynthetic stress, with moss F_v/F_m declining by a site average of 40% from low to high urban "class" (0.76 to 0.45). We also found increased wax deposition in moss with urbanisation, raising the hypothesis that it is a defence mechanism in these environments. Our study highlights the stressors potentially limiting moss persistence in cities and quantifies moss turf ability to trap PM. This trait could be exploited in applications relating to urban greening or air quality and should be further explored.

Keywords: urbanisation; air quality; phytoremediation; particulate matter; chlorophyll fluorescence

Presentation type: Oral Presentation

Bryophytes of Myanmar. Inventory, characteristics and affinities.

<u>Masanobu Higuchi¹</u>, Mu Mu Aung², Nobuyuki Tanaka¹

¹ Department of Botany, National Museum of Nature and Science, Tsukuba, Ibaraki 305-0005, Japan (higuchi@kahaku.go.jp); ² Forest Research Institute, Forest Department, Ministry of Natural Resources and Environmental Conservation, Yezin, Nay Pyi Taw, Myanmar

Indochina is bryologically one of the least known regions in Asia. Especially the flora of Myanmar (Burma) is still insufficiently known. It lies between latitudes 9° and 29°N, and longitudes 92° and 102°E, situating in the monsoon region of Asia. It also extends from the mangrove forests and coral reefs of the Andaman Islands in the south to the snow-capped peaks of Mt. Kakaboradzi (5881 m) in the north. The situation in the surrounding area suggests that there is incredible floral diversity in Myanmar. The National Museum of Nature and Science, Japan, together with the Forest Research Institute, Myanmar, embarked the international joint inventory research project to collect the materials for a "Flora of Myanmar" under the MoU since 2016. This study aims to present a preliminary report of the results of our field surveys, intending to highlight the characteristics and affinities of the bryophytes of Myanmar. Field surveys were conducted on Tanintharyi Nature Reserve (elevation lower than 250 m) of southern part in January, 2017, and on the montane area of Chin State (elevation 1200-2600 m) of western part in November, 2017. A total of ca. 550 bryophyte specimens were collected in both field surveys. The bryophytes recognized in Tanintharyi Nature Reserve mainly consist of the species of Calymperaceae, Fissidentaceae, Sematophyllaceae, Lejeuneaceae, etc., which are similar to those of lowlands of subtropical or tropical regions of Asia. On the other hand, the bryophytes recognized in montane area of Chin State are mainly composed of the species of Brachytheciaceae, Dicranaceae, Hylocomiaceae, Meteoriaceae, Neckeraceae, Thuidiaceae, Lepidoziaceae, Plagiochilaceae, etc., which are similar to those of the forests of temperate regions of Asia.

Keywords: Bryophytes, floristic element, inventory, Myanmar

Abstract Title: Knock-down of essential subunits of SCM5/6 complex in *Physcomitrella patens*

Marcela Holá, Radka Vágnerová, Karel J. Angelis

The Czech Academy of Sciences, Institute of Experimental Botany, Na Karlovce 1, CZ-16000, Prague, Czech Republic (hola@ueb.cas.cz)

SMC5/6 complex participates in many vital cell processes including DNA repair and most of its eight subunits are essential. In haploid Physcomitrella patens mutations of essential genes leading to lossof-function are lethal and their study requires rather attenuation of transcription or expression that allows partial viability of mutant than hard knock-out. We used two different techniques - RNA interference (RNAi) and CRISPR/dCas9 interference (CRISPRi) - to knock-down essential genes of SMC5/6 complex for generation of lines manifesting deficiency of protein of interest, but still viable. RNAi was performed by transformation of plasmid expressing short hairpin RNA (shRNA) targeted to SMC6 mRNA and produced stably transformed lines with decreased levels of SMC6 transcript and strong phenotype. Nevertheless, the RNAi effect disappeared after 3-4 cycles of subcultivation of primary transformants. The loss of SMC6 gene silencing was most probably caused by methylation and subsequent transcriptional inactivation of shRNA construct. To gain lines with stably silenced subunits of SMC5/6 complex, plasmid expressing catalytically dead Cas9 (dCas9) and gRNA targeted near to START codon of NSE1 or NSE4 was introduced into protoplasts of Physcomitrella. Recovered CRISPR/dCas9 lines were screened by qRT-PCR for transcript levels of NSE1 or NSE4 and the lines with decreased level of NSE1, NSE4 mRNA were tested for their sensitivity/resistance to acute DNA damage induced by radiomimetic drug bleomycin. Although only mild reduction (20-30%) of NSE1 and NSE4 mRNA levels was detected the CRISPRi lines manifested clear resistance to acute DNA damage. The decreased mRNA level and resistant phenotype has been maintained even during consequential cycles of subculturing. Our data show that, unlike RNAi, CRISPRi allows long-term effective repression of essential genes in Physcomitrella.

Supported by the Ministry of Education, Youth and Sports of the Czech Republic COST Action CA15132 (grant LTC17047/2017).

Keywords: RNA interference, CRISPR/dCas9 interference, SMC5/6 complex

Evolutionary conserved epigenetic silencer Polycomb repressive complex 2 in the bryophyte *Physcomitrella patens*

Helena Hönig Mondeková^{1,2}, <u>Kateřina Kabeláčová^{1,2}</u>, Katarina Landberg³, Eva Sundberg³, Iva Mozgová^{1,2}

¹ Institute of Plant Molecular Biology, Biology Centre of the Czech Academy of Sciences, 370 05 Ceske Budejovice, Czech Republic (katerina.kabelacova@umbr.cas.cz); ² Faculty of Science, University of South Bohemia, 37005 Ceske Budejovice, Czech Republic; ³ Department of Plant Biology and Forest Genetics, Uppsala BioCenter, Swedish University of Agricultural Sciences and Linnean Centre for Plant Biology, SE–750 07 Uppsala, Sweden

Polycomb Repressive Complex 2 (PRC2) is a protein complex composed of proteins that belong to the family of Polycomb group (PcG). Together with the Trithorax group (TrxG), PcG complexes regulate development and differentiation in all eukaryotic organisms. PRC2 was firstly described in Drosophila melanogaster and later on recognized as an evolutionarily conserved stabiliser of gene suppression. Here, we examine PRC2 subunits FERTILIZATION INDEPENDENT ENDOPLAST (FIE) and CURLY LEAF (CLF) in Physcomitrella patens to determine their effect on phototrophic development.

P. patens PRC2 mutants ($\Delta PpCLF$ and $\Delta PpFIE$) generated in our laboratory were analysed by RT-qPCR which showed a successful deletion of *PpCLF* and *PpFIE* gene correspondingly. Western Blot assay was further conducted to confirm the loss of post-transcriptional modifications which are installed by PRC2. A phenotypic analysis was performed to describe the defects in the mutants. To limit the developmental contribution of PRC2 absence to the analysed phenotypes, we focused especially on protonemal stage. PRC2 dysfunction in P. patens under long term cultivation in the presence of glucose results in morphologically different phenotype. Together with increased light intensity it promotes a formation of structures described as sporophyte-like bodies. Further RT-qPCR analysis of candidate genes whose dysregulation may be connected to the observed light and metabolism-related phenotypes provides first insights into the role of PRC2 in the moss photoautotrophic growth and development.

Keywords: Bryophyte epigenetics, PRC2 transgenic lines, *In vitro* cultivation, phototrophic growth and development

Interspecific variation in the fitness of six Iberian soil mosses assessed through biomass production and colonization success

<u>Fernando Hurtado^{1,2}</u>, Belén Estébanez², Manuel Molina^{1,2}, Cristina Ronquillo¹, Pedro Aragón¹, Joaquín Hortal¹, Nagore G. Medina²

¹ Department of Biogeography and Global Change, Museo Nacional de Ciencias Naturales (MNCN-CSIC), Madrid 28006, Spain(fhurtado@mncn.csic.es);

² Departamento de Biología (Botánica), Facultad de Ciencias, Universidad Autónoma de Madrid, Madrid 28049, Spain;

Vegetative dispersal is key for understanding the distribution of species with vegetative reproduction. However, the mechanisms behind it are often poorly understood. In moss species in which all species are potentially capable of propagating from fragments the role of mechanical fragmentation is crucial. We examined the colonization success of different fragment sizes and types in six Iberian soil mosses (Hypnum cupressiforme, Dicranum scoparium, Ptychostomum capillare, Homalothecium aureum, Tortella squarrosa and Syntrichia ruralis). We obtained three classes of shoot fragments through mechanical milling and sieving of dry samples (large, medium and small). Additionally, wet fragments were directly cultured as a fourth distinct class. All fragment classes were examined using light microscopy to assess fragment origin (leave or shoot), presence of meristems and size. After two months in culture, we measured the percentage of colonized surface and biomass (i.e. the viable biomass in relation to sown dry weight). Fragments milled in wet had, in general, lower success in our experiments. For dry fragments the intermediate size shows lower intraspecific variability and a fair performance (above 25% of colonization), while small fragments present much worst performance. Some species stand out because of their higher variability. Two of the species show larger differences between fragment sizes. H. cupressiforme had biomass production rates above 100% in the large fragments, and below 30% for the rest of fragment types, while P. capillare presents biomass production above 100% for intermediate and big fragment sizes and below 50% for the rest. These results seem to be related with the presence of a sufficient number of apical meristems in the shoot fragments and the ability of the species to reproduce directly from leaf cells. We also show that intermediate fragment sizes are probably more effective for culture experiments. Regarding natural population establishment success, it is interesting to note that the smallest fragment sizes, that have the higher potential for longer distance dispersal, show the lowest success rates.

Keywords: Bryophyte dispersal, colonization traits, fitness components, fragment viability, asexual reproduction.

Drivers of ground bryophytes diversity in temperate beech forests - stand structure vs. soil characteristics

Miloš Ilić¹, Ružica Igić¹, Mirjana Ćuk¹, Dragana Vukov¹

¹University of Novi Sad, Faculty of Sciences, Department of Biology and Ecology. Trg Dositeja Obradovića 2, 21000 Novi Sad, Serbia (<u>milos.ilic@dbe.uns.ac.rs</u>)

Bryophyte diversity and distribution is defined by large number of factors which act within time and space scale. Various studies showed the importance of soil characteristics and stand structure on bryophyte diversity and distribution. However, studies which deals with influence of these factors on ground bryophyte flora in temperate beech forests of South-eastern Europe are still lacking. The aim of this study was to determine the influence of soil characteristics and stand structure on bryophyte diversity in broadleaved temperate forests with Fagus sylvatica as dominant species. The survey was performed in Fruška gora Mountain (Northern Serbia) on 5 forest sites. Within each site, 5 plots (100x100 m in size) were placed, and Microcoenose sampling method was performed for quantitative sampling of ground bryophytes. Soil moisture, soil temperature and pH were measured within each site as soil characteristics, as well as herbaceous cover, litter cover, stream distance, number of trees and bushes as characteristics of stand structure. Shannon diversity index and Pielou's evenness index were used as diversity measures. Generalized linear model (GLM) and CCA analysis were performed for testing the influence of individual factors on bryophyte diversity. Variation partitioning analysis was performed for testing influence of stand structure and soil characteristics on bryophyte diversity. Total number of 54 species was found on all 5 sites. GLM analysis showed that all factors, except soil temperature and soil pH, have significant influence on ground bryophyte diversity. CCA analysis showed that largest percent of variability in bryophyte diversity can be explained by litter cover, herbaceous cover, soil moisture, tree number and stream distance, respectively. Variation partitioning analysis showed that stand structure explains 18.7% of total variability in bryophyte diversity, while soil characteristics explains 9.5%. Together, these two factors explain 14.1% of variability. The results of this study showed that stand structure is a more important factor influencing ground bryophytes in comparison to soil characteristics. Determination of the most important factors that influence bryophyte diversity in forest stands is of high importance in forest ecology, especially for establishing adequate methods of management in order to preserve biodiversity.

Keywords: ground bryophytes, forests, stand structure, beech

Morphological and phylogenetic notes on Trachycarpidium species (Pottiaceae, Bryophyta)

Yuya Inoue^{1,2}, Belén Estébanez Pérez³, Hiromi Tsubota¹

¹ Program of Basic Biology, Graduate School of Integrated Sciences for Life, Hiroshima University, 1-3-1 Kagamiyama, Higashi-hiroshima, Hiroshima, Japan (jujainoue@hiroshima-u.ac.jp); ² Hattori Botanical Laboratory, 6-1-26 Obi, Nichinan, Miyazaki, Japan; ³ Department of Biology, Faculty of Sciences, Universidad Autónoma de Madrid, Ciudad Universitaria de Cantoblanco, 28049 Madrid, Spain

Pottiaceae is the most generic- and species-rich family in Bryophyta (Mosses), with around 1500 species in ca. 80 genera. The taxonomy of the family has been difficult and controversial due to problems of morphological variability, the unclear significance of several anatomical characters, the reduced size of many species, the obscure leaf areolation, and sterility of many specimens. Although some molecular phylogenetic studies have been conducted to resolve relationships of the family at (supra-) generic level, the status and phylogenetic position of ca. 20 genera are still unrevealed due to the lack of useable specimens for successful DNA extraction and amplification. Trachycarpidium Broth. is one of these genera. It comprises five species with tropical distribution. This genus is characterized by immersed, cleistocarpous capsules with mamillose-bulging exothecial cells. In contrast to its unique sporophytic characters, the gametophytic similarity to Tortella (Müll. Hal.) Limpr. or Weissia Hedw. was suggested by some authors. However, the generic status of Tracycarpidium has been retained due to the lack of decisive evidence, as molecular data were not available then. In the present study, we investigated the anatomical structure of capsules in Trachycarpidium. In the context of previous molecular studies, here we also successfully obtained chloroplast DNA sequences from two species of the genus, and inferred their phylogenetic positions within the Pottiaceae. The anatomical comparison of the capsules allowed us recognizing two different thickening types of exothecial cell walls (outer walls): 1) evenly thick, including both sides and tip of its central mammilla; 2) markedly thicker at the mamilla sides and much thinner at its tip. We suggest that thickenings of exothecial walls could be useful as a taxonomic character in this group for species discrimination. However, phylogenetic analyses using concatenated sequences of chloroplast rbcL and rps4 genes showed that T. brisbanicum (Müll.Hal.) I.G.Stone and T. lonchophyllum (G.Roth) R.H.Zander were nested in the Weissia clade, pointing at a polyphyletic origin for capsules with mamillose-bulging exothecial cells. Our results support the congeneric treatment of Weissia and Trachycarpidium and also suggest that the sporophyte morphological diversity is much higher than expected within Weissia.

Keywords: Pottiaceae, Trachycarpidium, Weissia, sporophyte anatomy, phylogeny

Towards a new IUCN based Red List of bryophytes for Switzerland

<u>Thomas Kiebacher</u>¹, Markus Meier¹, Julie Steffen¹, Norbert Schnyder¹, Niklaus Müller¹, Ariel Bergamini², Edi Urmi¹ & Heike Hofmann¹

¹ Swissbryophytes – National Data and Information Centre for Swiss Bryophytes, Department of Systematic and Evolutionary Botany, University of Zürich UZH, Switzerland (thomas.kiebacher@uzh.ch)

² Swiss Federal Institute for Forest, Snow and Landscape Research WSL; Birmensdorf, Switzerland

At the Swiss Data and Information Centre for bryophytes work on a new Red List is underway. Although the data basis is extensive (> 300'000 data sets) various challenges have to be faced: heterogeneity of data, taxonomic changes, interpretation of IUCN criteria as well as an appropriate assessment of global and regional threats such as climate change and landscape changes. Specific field surveys (e.g. searching for rare species) and herbarium revisions are carried out to close data gaps. Furthermore, with the intention to provide an improved data basis for future red lists, a permanent plot survey is set up to monitor selected priority species and habitats. The new Red List is to be published in 2020.

Keywords: Climate change, Conservation, Permanent plots

Presentation type: Poster Presentation

Specificity of Caucasian liverwort flora: phytogeographical peculiarities and hidden diversity

<u>Nadezhda A. Konstantinova¹, Anna A.Vilnet¹</u>

¹ Polar-Alpine Botanical Garden-Institute of the Kola SCI Center of Russian Academy of SCI, Kirovsk, Murmansk Province, RUSSIA (nadya50@list.ru)

We have analyzed the liverwort flora of Caucasus based on data obtained during the study of liverworts diversity in this mountains for ca. 15 years. Ca 4000 specimens from almost sea level up to 3000 m alt. were gathered mainly in Russian part of the Caucasus and about 2000 of them were identified. Labels data are available in Cryptogamic Russian Information System (http://kpabg.ru/cris/?q=node/16). Some of the specimens were studied recently using molecular phylogenetic approach. Many new species for the Caucasus and some new ones for science were revealed. At present ca. 200 species are recorded for the Russian part of the Caucasus. The core of the flora are widespread Holarctic liverworts including boreal and arctic-montane species restricted mostly to the middle and upper belts of the Caucasus that were classified by A.Takhtadjan (1978) as the Caucasian Province of Circumboreal Region. The arctic-montane species are restricted to upper belts of Caucasus and probably are remnants of arctic-tertial flora. A number of predominantly Mediterranean species are in Caucasus near the eastern and northern limits of their distribution and occur mostly in presumably not glaciated Euxine Province. Several mostly Asian species are in the Caucasus on their western limit. The long isolation of the Caucasus vegetation is reflected in its richness in endemic flora of vascular plants whereas among hepatics for a long time only Solenostoma caucasicum (Váňa) Konstant. was known as endemic of the Caucasus. Using integrative approach, we found that some species are represented in the Caucasus by distinct genetic units. Some of them were described as new for science, e.g. Jubula hutchinsiae (Hook.) Dumort. subsp. caucasica Konstant. & Vilnet., Jungermannia calcicola Konstant. et Vilnet. We found as well that Caucasian population of Liochlaena subulata (A.Evans) Schljakov is molecularly distinct from that in Europe and Siberia. One more Jungermannia species presumably new for science and phylogenetically allied to J. atrovirens, was discovered by us in Euxine Province of the Caucasus and will be described in nearest time. We believe that based on integrative approach careful study of recent Caucasian liverworts collections will show relatively high level of hidden endemism of the Caucasian liverwort flora.

Keywords: Liverworts diversity, phytogeography, hidden endemism, the Caucasus

A multi-scale approach to drought in *Sphagnum*, the cellular basis of an ecosystem engineer

<u>Willem Q.M. van de Koot¹</u>, Ying Wang¹, Candida Nibau¹, John H. Doonan¹

¹ Institute of Biological, Environmental and Rural Sciences, Aberystwyth University, Plas Gogerddan, SY23 3EB, Wales (U.K.) (wqv@aber.ac.uk).

As a key component of most peat-forming ecosystems, Sphagnum is of major importance for the ecosystem services it provides. Due to the increase of global temperature and extreme weather events, in combination with anthropogenic alteration of the ecosystem, the peatlands are increasingly exposed to oxygen and as the length of dry periods increases, the peat deteriorates and releases large amounts of greenhouse gases. Therefore, understanding the adaptations that these mosses have for avoiding or coping with desiccation is essential for the preservation of functioning peatland ecosystems. By combining approaches at the ecological, photo-physiological and genetic levels, the present research aims to elucidate the basic processes that are involved in desiccation tolerance in *Sphagnum*, such as the typical distinction that is made between hollow and hummock occupying species and their varying degrees of desiccation tolerance. Through the measurement and imaging of photo-physiological variables in different ecological settings such as stem density and subjection to periodic drought. Initial results regarding the effect of colony density on photophysiological parameters suggest that at higher stem density, S. fallax can tolerate longer periods of drought and maintains photosynthetic activity in the lower parts of the plant. Future aspects of this project will replicate these experiments on different species of Sphagnum, and also investigate possible within-species variation as a result of ecological niche differences. In parallel, we aim to look at the expression levels of desiccation-related genes and how their expression changes in different tissues, sections of the plant and throughout time during the drought and recovery period. Taken together our results will provide a better understanding of the genetic and physiological basis of desiccation tolerance in Sphagnum and will help inform peatland recovery projects.

Unique epigenetic features of ribosomal RNA genes (rDNA) in early diverging plants (bryophytes)

<u>Alena Kuderová¹</u>, Roman Matyášek¹, Alice Krumpolcová¹, Jana Lunerová¹, Eva Mikulášková², Josep A. Rosselló³, Aleš Kovařík¹

¹Department of Molecular Epigenetics, Institute of Biophysics, Academy of Sciences of the Czech Republic, v.v.i., Královopolská 135, 612 65 Brno, Czech Republic (<u>alenakud@ibp.cz</u>, kovarik@ibp.cz); ²Department of Botany and Zoology, Masaryk University, Kotlarska 2, 61137 Brno, Czech Republic; ³Jardín Botánico, ICBiBE-Unidad Asociada CSIC, Universidad de Valencia, c/Quart 80, E-46008, Valencia, Spain

Introduction: In plants, the multicopy genes encoding ribosomal RNA (rDNA) typically exhibit heterochromatic features and high level of DNA methylation. Here, we explored rDNA methylation in Bryophyta (15 species) and Marchantiophyta (4 species) using methylation-sensitive Southern blot hybridization in all species. We also carried out whole genomic bisulfite sequencing in *Polytrichum formosum* and *Dicranum scoparium* and used available model plant methyloms in *Physcomitrella patents* and *Marchantia polymorpha* to determine rDNA unit-wide methylation patterns. Chromatin structure was analyzed using *in situ* hybridization and immunoprecipitation assays.

Results: In contrast to seed plants, bryophytes rDNAs were efficiently digested with methylationsensitive enzymes indicating no or low levels of CG and CHG methylation in these loci. The rDNA methylom analyses revealed variation between species ranging from negligible (<3%, *P. formosum*, *P. patens*) to moderate (7 and 17% in *M. polymorpha* and *D. scoparium*, respectively) methylation levels. There were no differences between coding and non-coding parts of rDNA units and between gametophyte and sporophyte tissues. However, major satellite repeat and transposable elements were heavily methylated in *P. formosum* and *D. scoparium*. In *P. formosum* rDNA, the euchromatic H3K4m3 and heterochromatic H3K9m2 histone marks were nearly balanced contrasting the angiosperms data where H3K9m2 typically dominates rDNA chromatin. In moss interphase nuclei, rDNA was localized at the nucleolar periphery and its condensation level was high.

Conclusions: Unlike seeds plants, the rRNA genes seem to escape global methylation machinery in bryophytes. Distinct epigenetic features may be related to rDNA expression and the physiology of these early diverging plants which exist in haploid state for most of their life cycles.

Keywords: rDNA, cytosine methylation, bryophytes, epigenetics, histone marks

Peptides encoded by long non-coding RNAs influence proteome in the moss *Physcomitrella patens*

<u>Anna Mamaeva¹</u>, Andrey Knyazev¹, Vassili Lazarev^{2,3}, Daria Kharlampieva², Victor Zgoda⁴, Nina Golub¹, Vadim Ivanov¹ and Igor Fesenko¹

¹ Shemyakin-Ovchinnikov Institute of Bioorganic Chemistry Russian Academy of Sciences, Laboratory of plant functional genomics and proteomics, Miklukho-Maklaya, 16/10, 117997, Moscow, Russian Federation (<u>AnnetteSt@yandex.ru</u>)

² Federal Research and Clinical Centre of Physical-Chemical Medicine, Moscow, Russian Federation;

³ Moscow Institute of Physics and Technology, Dolgoprudny, Moscow region, Russia;

⁴ Laboratory of System Biology, Institute of Biomedical Chemistry, Moscow, Russian Federation.

The biological functions of peptides encoded by small Open Reading Frames (sORFs) are generally unknown. However, based on few studied peptides, we can suggest that they can regulate the diverse range of cellular processes in plants, such as cell proliferation (ROT4), nodulation and sugar metabolism (ENOD40), pollen grains germination (Zm908), programmed cell death (KOD) and root growth (PLS).

Using mass spectrometry analysis, we identified nine translatable sORFs located on long non-coding RNAs in the moss *Physcomitrella patens*. To study an impact of sORF-encoded peptides (SEPs) on plant growth and development, four of these SEPs (PSEP1, PSEP3, PSEP18 and PSEP25), ranged from 40 to 61 aa, were chosen for knockout by CRISPR-Cas9 method and overexpression in *P. patens*. Phenotypic analysis of mutant lines have shown that the knockout of all SEPs led to significant reduce in growth rate of moss protonemata. However, the number of leafy shoots was increased in PSEP25 knockout lines.

To evaluate the changes at the protein level, we performed quantitative proteomic analysis with iTRAQ labels. Proteomic analysis of SEPs knockout lines revealed the different number of differentially expressed proteins (DEPs). Proteomic changes in PSEP1 and PSEP3 (142 differentially expressed proteins in both knockout lines) are more extensive than in PSEP18 and PSEP25 (8 and 14 differentially expressed proteins respectively) knockout lines. According to GO enrichment analysis both PSEP1 and PSEP3 knockout lines have shown a similar down-regulation of proteins participated in basic metabolic processes. Also, knockout of PSEP1 led to down-regulation of proteins participated in translation, whereas PSEP3 knockout led to changes in photosynthesis and photorespiration processes. Both PSEP18 and PSEP25 knockout induce changes in content only of a small number of individual proteins. Taken together, morphological and proteomic analysis of SEPs knockout lines have shown that peptides PSEP1, 3, 18 and 25 can regulate growth and development of moss plants. Thus, peptides encoded by long non-coding RNAs play an important role in plants.

This work was supported by grant № 17-14-01189 from Russian Scientific Foundation.

Keywords: proteomics, functional peptides, Physcomitrella patens

Towards a clarification of the Aloina catillum (Müll. Hal.) Broth. (Pottiaceae) complex

Mónica Martínez, M. Teresa Gallego, Juan A. Jiménez, Juan Guerra, María J. Cano.

Área de Botánica, Departamento de Biología Vegetal, Universidad de Murcia, Spain

(monica.martinez11@um.es; mcano@um.es)

Aloing Kindb. (Pottiaceae) is a rather widespread genus found on most continents, which comprises 12 species. This genus has been infrequently collected in South America where only five species are recognized. One of these species, Aloina catillum (Müll. Hal.) Broth. is endemic of the Tropical Andes. It is mainly characterized by suborbicular leaves with apex broadly cucullate, hyaline marginal cells at the leaf base, costa with 2-3 stereids layers, red seta, ovoidcylindrical and dark red coloured capsules, peristome strongly twisted, with basal membrane differentiated and operculum conical. However, these characters can overlap with those of A. rigida (Hedw.) Limpr. and especially A. brevirostris (Hook. & Grev.) Kindb. In this study we aim to evaluate the phylogenetic relationships and to critically examine boundaries between these three species that are easily confused with each other based on their similar morphologies. To this purpose a morphological study and a molecular analysis using the rDNA ITS 1-2 region in several specimens of South American species of Aloina was carried out. In addition, other species of the genus such as A. brevirostris, A. bifrons (De Not.) Delgad., A. aloides (W.D.J. Koch ex Schultz) Kindb. and A. ambigua (Bruch & Schimp.) Limpr. from other locations worldwide were included. Our phylogenetic results indicate that A. catillum specimens constitute several groups nested in the clade of the genus suggesting different species, while the morphological analysis does not show a clear variation among these entities. The controversial data obtained show the necessity to study more samples and the use of other molecular markers in order to clarify the possibility of the existence of cryptic species.

Keywords: Aloina, molecular phylogeny, South America, taxonomy.

Interaction between three Mediterranean mosses: influence of competition and abiotic factors in biogeographical distribution.

Paula Martínez Domínguez¹, Belén Estébanez¹ & María Leo²

- 1. Comisión Docente de Botánica, Dpto. Biología, Fac. Ciencias, UAM, Madrid, Spain (paulamdguez1@gmail.com, belen.estebanez@gmail.com)
- Dpto. Biodiversidad, Real Jardín Botánico, (RJB-CSIC), Madrid, Spain. (<u>maria.leomontes@gmail.com</u>)

The distribution and dynamics of plant communities depend on abiotic factors and biotic interactions. Aiming at assessing the role of these factors on the presence, coexistence and distribution of moss species, here we present a study assessing the effect of interspecific competition along an altitudinal gradient (500 m-1500 m). We have tested three species, Homalothecium aureum, Hypnum cupressiforme and Tortella squarrosa, all very common in central Spain. We have used two experimental approaches, one based on natural conditions and the other on artificial outdoors settings. In each, we established three different altitudinal bands. In the natural habitats, we have registered the abundance and coexistence through systematic samplings favouring plots where coexistence occurs. In the outdoor cultures we have established artificial moss communities using individual shoots to obtain homogeneous tufts, both monospecific and mixed (using all three species, or in pairs), grew them settings, and assessed their interactions through differences in the moss area coverage, dry weight and shoot length for each species. In their natural environment, we have not observed all three species coexisting. H. aureum is present along the whole altitudinal gradient. T. squarrosa predominates in the lowest and medium altitudinal band, and H. cupressiforme is dominant in the highest localities. However, when coexisting with the other species, *H. aureum* seems to perform better. In outdoor cultures, all three species grow better in the plot at lowest altitude. Mixed communities of two species and, to a lesser extent, the monospecific ones, show the largest increase of area coverage. Both T. squarrosa and H. cupressiforme are able to survive outside their natural altitudinal range. Mixed communities of all three species seem to perform worse, as they only show a coverage increase in the lowest altitude. H. cupressiforme presents the highest growth, although *H. aureum* is visually the healthiest. We conclude that climatic variables matter but interspecific interactions affect greatly the performance of individual species, highlighting the importance of considering them in ecology studies.

Keywords: interspecific interactions, coexistence, altitudinal gradient, outdoor cultures.

Presentation type: Oral Presentation, <u>Poster Presentation</u> **Dynamics of development and change of biodiversity in natural forests**

Mikulášková E., Běták J., Halda J., Procházka J.

Výzkumný ústav Silva Taroucy pro krajinu a okrasné zahradnictví, v.v.i., Odbor ekologie lesa, Lidická 25/27, CZ-602 00 Brno (evamikul@gmail.com)

Natural forests have been transformed during centuries into a forest-free area or cultural forests with different degree of conservation in Central Europe. If we want to support a primaeval character of cultural forests by management, we need to understand the processes and dynamics in the natural forests. We investigate species diversity of epiphytic and epixylic bryophytes in several types of primaeval forests. Based on long-term dendrometric measurements, species composition is possible to link to clearly identified objects (live and dead trees, stumps, rotting logs at all stages of decomposition). It provides information about distribution and ecology of species and real-time community development, it allows to study interspecific and ecological relationships across groups of organisms and to study communities bounded to less common tree species (fir, elm, oak).

We collect species data on permanent circular plots of 500 m2.

In 2018, research was focused on two forest reserves: Stožec (Šumava NP) and Rejvíz (Jeseníky PLA). The Stožec research area includes mixed debris forests with large amounts of old tree stands and abundant decaying wood, including logs with a large diameter. The vegetation in the Rejvíz research area includes pine and spruce peat forests with a low amount of decaying wood. Over 400 objects were examined in both reservations.

113 species of epiphytic and epixylic bryophytes including wood specialists were recorded in the Stozec reserve, of which 14 species are included in one of the categories of the red list of bryophytes of the Czech Republic (Kucera et al. 2012). 59 species of bryophytes were recorded in the Rejvíz research area, of which 6 species are included in the red list of the Czech Republic. The presence of rare species on the site, including the highly endangered ones, confirms that existing management is appropriate for the study group. The non-intervention mode, along with leaving dead wood on the site, provides the necessary substrate for species typical of natural forests.

Kučera J., Váňa J. & Hradílek Z. (2012): Bryophyte flora of the Czech Republic: updated checklist and Red List and a brief analysis. – Preslia 84: 813–850.

Keywords: epiphytic and epixylic bryophytes, primaeval forests, biodiversity

Can the content of bryophyte ultraviolet-absorbing compounds be estimated by non-destructive methods? A case study in the Colombian Andes

Laura Monforte¹, Luis-Carlos Montenegro², Edgar Linares², Jaime Uribe², Javier Martínez-Abaigar¹, Encarnación Núñez-Olivera¹

¹ Facultad de Ciencia y Tecnología, Universidad de La Rioja, 26005 Logroño (La Rioja), Spain (<u>laura.monforte@unirioja.es</u>); ² Departamento de Biología, Universidad Nacional de Colombia, Bogotá, Colombia

Bryophytes inhabiting high mountains like the Andes are exposed to harsh environmental conditions, such as high levels of ultraviolet (UV) radiation. The bryophyte UV protection capacity is usually evaluated by quantifying their amount of phenolic UV-absorbing compounds (PUVAC) by spectrophotometry or HPLC. Unfortunately, these methods are destructive and not applicable under field conditions. However, Dualex[®] (Force-A) is a portable instrument allowing the non-destructive estimation of PUVAC in plants, although it has rarely been tested (if ever) on bryophytes. In this study, we measured the bulk level of PUVAC in 56 samples of 28 bryophyte species (18 mosses, 9 liverworts and 1 hornwort) by both destructive (methanolic extracts and then spectrophotometry) methods in the laboratory and non-destructive (Dualex®) methods in the field. Our aim was to evaluate the potential use of Dualex® to assess the UV protection capacity of bryophytes under field conditions. The experiment took place in the Colombian Andes in July 2018 and bryophytes were collected in an elevation range of 700-3500 m under different sun exposures, from full sun to deep shade. For the spectrophotometrical measurements, we differentiated the PUVAC present in vacuolar soluble forms (S-PUVAC) from those present in cell wall-bound insoluble forms (I-PUVAC), because each type of compounds may represent a different modality of UV protection. We also calculated the total PUVAC (T-PUVAC) as the sum of S-PUVAC and I-PUVAC. Regarding Dualex® measurements, we obtained both the flavonol index (Flav) and the anthocyanin index (Anth), given that flavonols and anthocyanins belong to different PUVAC families. Mosses showed higher contents of I-PUVAC and T-PUVAC than liverworts, whereas liverworts showed higher contents of S-PUVAC than mosses, as occurred in temperate bryophytes. The only hornwort collected showed higher contents of T-PUVAC and S-PUVAC than mosses and liverworts, respectively, and similar I-PUVAC contents to liverworts. Flav (but not Anth) reproduced the same results of T-PUVAC for mosses and liverworts. In addition, Flav was strongly and positively correlated with I-PUVAC, in both mosses and liverworts, but no correlation was found between Flav and S-PUVAC. Thus, Dualex® have certain limitations to be used in the non-destructive evaluation of UV protection in bryophytes.

Keywords: Ultraviolet protection, Dualex®, mountain bryophytes, phenolic compounds, flavonoids

The Moss Flora of Nuluhon Trusmadi Forest Reserve in Sabah, Malaysian Borneo.

Andi Maryani A Mustapeng¹, John B Sugau¹, Monica Suleiman²

¹ Forest Research Centre, Sabah Forestry Department, P.O. Box 1407, 90715 Sandakan, Sabah, Malaysia (andimaryani.mustapeng@gmail.com); ² Institute for Tropical Biology & Conservation, Universiti Malaysia Sabah, Jalan UMS, 88400 Kota Kinabalu, Sabah, Malaysia.

Nuluhon Trusmadi Forest Reserve (NTFR) is located in the central part of Sabah, Malaysian Borneo, and covers an area of 74,736 ha. NTFR is gazetted as Protection Forest Class I, where timber-harvesting activities are not permitted since 2010. The main aim of this research is to carry out a comprehensive study on the moss flora of NTFR, to collect, identify, prepare descriptions, construct keys to species, and to assess the general phytogeographical affinities of mosses from NTFR. Specimens of mosses were randomly collected in all existing trails and excursions in accessible areas of the reserve. Herbarium specimens from SAN and BORH are also included in this study. In total, 903 moss specimens were collected from the reserve and were examined. From this collection, 232 species, 18 varieties and five subspecies belonging to 95 genera and 34 families of mosses have been identified. This number represents 33.2% of the 767 reported mosses from Borneo and 39.4% of the 647 mosses in Sabah. Sematophyllaceae appeared to be the most dominant family in NTFR, with 23 species and seven varieties, followed by Calymperaceae with 27 species, and Pylaisiadelphaceae with 19 species and two varieties. Out of these, 16 taxa are newly reported for Sabah including 11 taxa new to Borneo. The moss flora of NTFR has a close affinity to Crocker Range Park (CRP), whereby 165 (55.6%) taxa of mosses in 81 (76.8%) genera and 30 (83.3%) families are common in both areas. NTFR also recorded four endemic species to Borneo, adding value to this forest reserve. In conclusion, NTFR has a relatively rich moss flora in terms of number of taxa, which is greater than Mount Tambuyukon but lower than Crocker Range Park in Sabah. There are moss entities in NTFR that are not found elsewhere in Borneo, including Mount Kinabalu, adding to the importance of protecting this area. As the current study only covered the accessible routes in this forest reserve, it should be further explored bryologically in the future, which will definitely increase the number of mosses presented here.

Keywords: Bryophytes, endemic, Mount Trus Madi, new records

Methods to Transform the New Model Species for Hornworts, Anthoceros agrestis

<u>Anna Neubauer¹</u>, <u>Manuel Waller¹</u>, Eftychios Frangedakis², Juan Carlos Villarreal³, Fay-Wei Li⁴, Peter Szovenyi¹

1 Department of Evolutionary and Systematic Botany, University of Zurich, Switzerland (anna.neubauer@uzh.ch, manuel.waller@uzh.ch, peter.szoevenyi@uzh.ch); 2 Department of Plant Sciences, University of Cambridge, United Kingdom; 3 Department of Biology, Laval University, Canada; 4 Boyce Thompson Institute for Plant Research, Cornell University, USA

Recently, a new bryophyte model species has received increased attention – the hornwort. Analyzing the hornwort genome will help to resolve fundamental questions of plant evolution and developmental biology, in general, and unique aspects of hornwort biology, in particular. However, having a fully sequenced genome does not fulfill all requirements of a well-established model organism. Here we report our progress concerning culturing, genetic transformation and laser capture microscopy assisted small-scale gene expression profiling (LCM RNA-seq) of the model organism *Anthoceros agrestis* achieved over the last year. In particular, we will elaborate on the tractability of the system for experimental research by first introducing main features of its nuclear genome and present our results on the protoplastation, transient transformation, and regeneration of hornwort tissues. Finally, we will show some of our first results on the stable genetic transformation of the species. Altogether, these achievements show that *Anthoceros agrestis* is becoming, step by step, a better-studied model organism for developmental, molecular, genomic, and genetic studies.

Keywords: Protoplastation, Transient Transformation, Stable Transformation, Hornwort Model Organism, *Anthoceros agrestis*

Revisiting the Mediterranean *Bartramia rosamrosiae* with phylogenetic, morphological and ecological tools

Konstantin Neumann¹, Jesús Muñoz² & Dietmar Quandt¹

¹ Nees-Institute for Biodiversity of Plants, University of Bonn, 53111 Bonn, Germany

(kneumann@uni-bonn.de)

² Real Jardín Botánico, CSIC, Plaza de Murillo 2, E-28014 Madrid, Spain.

Species delimitation in mosses is not a trivial task in many cases. Their often rather small size and limited amount of prominent diagnostic structures make it hard to pinpoint critical identification criteria between closely related species. This is also true for the section Strictidium of the acrocarpous moss family Bartramiaceae and its prominent widespread "Mediterranean" species Bartramia stricta and relatives. Based on detailed molecular and morphological analyses, Damayanti et al. (2012) recently showed that B. stricta from South America is indeed a different species compared to the Mediterranean B. stricta from Europe and California. As the Bridel type refers the South American species Damayanti et al. (2012) segregated the "Mediterranean" species from B. stricta as a new species *B. rosamrosiae*. But in 2014 Müller claimed that this species was already validly described as *B. aprica* from the Sudan and thus rendering *B. rosamrosiae*.as an illegitimate name. Since no samples from the Sudan were incorporated in the former study by Damayanti et al. (2012) and the type pictures provided by Müllers (2014) are somehow of intermediate morphology we conducted a more detailed study on B. aprica. Here we show that B. rosamrosiae is a different species from *B. aprica*, by providing molecular as well as morphological and ecological evidence. The molecular analyses clearly show a significantly supported sister group relationship of B. aprica and B. rosamrosiae, each being monophyletic. Furthermore the morphological analysis shows that B. rosamrosiae has an overall higher plant length and smaller stem diameter as well as a slimmer central cylinder diameter, in comparison to *B. aprica*. In addition climate niche modelling shows that B. aprica prefers much more arid and desert like habitats, while B. rosamrosiae occupies area with Mediterranean climate. Our results show, that species delimitation in mosses should sometimes be done with more care, a sensible combination of tools and looking at a representative set of vouchers.

Keywords: Bartramiaceae, climate niche modeling, species delimitation, ecology, morphology

Liverwort richness and range size distribution along elevational gradient in Serbia

Jovana P. Pantović¹, Nada M. Nikolić², Marko S. Sabovljević¹

¹Institute of Botany and Botanical Garden, Faculty of Biology, University of Belgrade, Studentski Trg 16, 11000 Belgrade, Serbia (<u>jpantovic@bio.bg.ac.rs</u>; marko@bio.bg.ac.rs); ²University of Marburg, Faculty of Geography, Ecological Plant Geography, Deutschhausstrasse 10, 35032 Marburg, Germany (nada.nikolic@geo.uni-marburg.de)

Biogeographical variation in species richness is important for our understanding and conservation of biological diversity. While there are insights of the tracheophyte's distribution along the elevational gradient in the Central Balkan, there are no similar studies on bryophytes in this region up to date. The aim of our research is to investigate how liverwort species richness varies along the elevational gradient in Serbia, and to evaluate if the range size of the species differs with elevation in the studied area. The study was conducted along a 2155 m altitudinal gradient in Serbia, a country in South-Eastern Europe. Digitized distribution data compiled from the literature and herbarium collections were used for the analysis. The presence of the species was interpolated between the minimum and maximum recorded elevations, and assessment of species richness for 100 m elevation bands is given. Elevational range size and the range midpoint is calculated for each taxon recorded. For species recorded at only one site, the adjusted range size of 100m was used. Species were considered wide-ranging in cases where their ranges cover at least half of the domain (range size \geq 1077m) and narrow-ranging if their ranges cover less than half the domain (range size < 1077). A humped relationship between species richness and elevation was observed, with maximum richness recorded at middle elevations, in 1100-1200 and 1200-1300m bands. According to the size of the occupied domain, the majority of the liverwort species in Serbia (56.5%) are narrow-ranged. Moreover, narrow-ranged liverworts tend to be more common towards the lower and higher elevations, whereas wide-ranging species occupy middle elevations. Only five wide-ranging species were found to extend their elevational ranges from the lowlands to the highest peaks. Our study provides the very first insights in liverwort's distribution and richness along elevation in Serbia. However, additional field data and more detailed research on influencing factors would lead to a better understanding of the distribution patterns of this plant group in Serbia.

Keywords: biogeography, elevation, liverworts, narrow-ranged species, wide-ranged species

Presentation type: Poster Presentation

Is drought stress driving the morphological variation of *Hypnum cupressiforme* HEDW. (Hypnaceae)?

Verena Pietzsch¹, Sonja M. Thielen¹, Martin Ebner¹

¹ Department of Geosciences, Eberhard Karls University of Tübingen, Hölderlinstraße 12, 72074 Tübingen (verena.pietzsch@uni-tuebingen.de)

Being poikilohydric plants, mosses depend on external capillary water to regulate their metabolic activities. The dependence on atmospheric conditions such as relative air humidity and air temperature led to the development of various adaptations to enable the survival of drought periods of different lengths. Drought stress is a real issue regarding the growth form and life form of, especially epiphytic, bryophytes. We hypothesize that drought stress is a driving factor for morphological variations in *Hypnum cupressiforme*.

The objective of this study is to investigate the morphological variation of *H. cupressiforme* within a tree and to check for a correlation to microclimatic conditions. Moss samples were taken at the 20th of October 2018 in two oak trees at the Hardtwald, Tübingen, Germany. The in total 39 samples are patches of about 10 x 10 cm and were collected from different positions within the tree from the stem basis to the treetop, including various inner and outer canopy positions. Every sample was first examined for species composition with the result of a definite dominance of *H. cupressiforme*. Thereby, distinct morphologies of this species have been observed depending on its origin. Climate data was collected over a two-month period from 21st of October to 17th of December 2018. Ten temperature/humidity (°C/%) data loggers were placed at several positions where moss samples have been taken within one of the two oak trees. For statistical analyses mean daily minimum relative humidity, mean daily maximum temperature and mean daily maximum VPD, as well as the number of days in which RH was below 85% have been calculated. To test for a correlation between morphology and microclimate point-biserial analyses have been performed. The results show a correlation between the morphology and the mean daily minimum air humidity, as well as the mean daily maximum VPD, whereas temperature has no effect.

Keywords: epiphytes, morphological variation, canopy microclimate

The rare and scattered moss *Acaulon piligerum* (Pottiaceae) new to the Iberian Peninsula with comparison to other Southern Hemisphere species of subgenus *Alaticosta*

Felisa Puche^{1,2}, José Gabriel Segarra-Moragues^{1,3}, Josephine Milne⁴

¹ Departamento de Botánica y Geología, Universidad de Valencia, Burjassot (Valencia), E-46100, Spain; ² (m.f.puche@uv.es); ³ (j.gabriel.segarra@uv.es).

⁴ Royal Botanic Gardens Victoria, Private Bag 2000, South Yarra, Victoria 3141, Australia. (josephine.milne@rbg.vic.gov.au).

Phascum bulbosum Voit. var. *piligerum* De Not. was described in 1938 from a sample collected in Cagliari (Sardinia) and five decades later transferred to *Acaulon (A. piligerum* (De Not.) Limpr.). After a revision of the type material, Sérgio in 1992 was the first to notice the presence of short lamellar outgrows on the ventral part of the nerve. This unique trait among Holarctic species served to assign it to subgenus *Alaticosta* I.G. Stone, together with the Australian *Acaulon chrysacanthum* I.G. Stone and the Australian-South African *A. leucochaete* I.G. Stone. The two southern hemisphere species have not been to date critically compared to *A. piligerum*, because very few collections have been made since its description, and some were misidentifications for the widespread *A. triquetrum* (Spruce) Müll.Hall.

In March 2017, during field work in temporary lagoon areas of Castilla la Mancha (Central Spain), samples of a moss which keyed to *Acaulon piligerum* were collected. The Spanish samples were studied and described in detail using optical and scanning electron microscopy techniques and compared to representative Australian specimens of subgenus *Alaticosta*.

Acaulon piligerum is a small, yellowish-brown, bulbiform plant, of 2.5-2.8 mm high that grows in compact turfs. Leaves are concave with nerve excurrent in a flexuose hyaline arista of 286-551 μ m long; cells on ventral side of costa smooth, inflated, with up to 2 chlorophyllose lamellae of up to 6 cells high that extend to the tip from the upper half of leaves. It is distinguished from *A. chrysacanthum* by its white flexuose arista of perichaetial leaves (straight, golden reddish in *A. chrysacanthum*). It is more similar to *A. leucochaete* with slight differences on leaf margin denticulation and papillosity of terminal cells of lamellae (both smooth in *A. piligerum*). Molecular data would be necessary to confirm the identity of these taxa. In its current circumscription *A. piligerum* has a scattered Mediterranean distribution including isolated localities in Sardinia, Crete, Lacroma Island and the new Spanish population reported here. As for its conservation status, according to these data it is proposed as Endangered (EN) B2a + biii.

Acknowledgments: We acknowledge financial support by the Ministry of Economy industry and Competitiveness (project CGL2016-80418-P).

Keywords: Disjunctions, Endangered species, Pottiaceae, Taxonomy

Climatic differences in the geographic range of moss sister taxa at both sides of the Atlantic.

<u>Cristina Ronquillo</u>¹, Jennifer Morales-Barbero¹, Pedro Aragón¹, Vicente Mazimpaka², Manuel Molina-Bustamante², Nagore G. Medina², Joaquín Hortal^{1,3}.

¹Department of Biogeography and Global Change, Museo Nacional de Ciencias Naturales (MNCN-CSIC), Madrid, Spain (cristinaronquillo@mncn.csic.es); ² Departamento de Biología (Botánica), Facultad de Ciencias, Universidad Autónoma de Madrid, Spain; ³ Centre for Ecology, Evolution and Environmental Changes (cE3c), Faculdade de Ciências, Universidade de Lisboa, Portugal

Bryophyte disjunctions between the New World and the Old World are common and often entail morphological variations that are structured in space. These geographically structured differences lead to controversies regarding the taxonomic status of the populations at both sides of the Atlantic. Those disjunctions have often been evaluated through genetic and/or morphological analyses, however, the ecological differences among disjunct mosses have rarely been assessed quantitatively, despite their potential importance for speciation processes. We evaluate the similarities on the climatic responses between two pairs of terricolous mosses under controversial taxonomic status: Homalothecium aureum and the synonymized H. pinnatifidum; and Pleurochaete squarrosa (=Tortella squarrosa) and the American P. luteola, so far considered synonymous of the former. We first modelled their responses to large-scale environmental gradients using publicly available data of their global distributions. Then, we evaluate the reliability of the modelling approach based on climatic information and Ecological Niche Factor Analysis (ENFA). Finally, we assess the similarities in their responses using niche overlap analysis. The results indicate that both moss taxa pairs show climatic differentiation. The Homalothecium pair shows similar responses to climate in both regions, but the American populations use a subset of the climatic domain occupied by the European populations. This contrasts with the *Pleurochaete* pair, which presents partially different distributions in some climatic gradients. Although the two taxa of this latter pair mainly respond to the mean temperature in the dry season, the European populations of *P. squarrosa* grow in areas that are colder during the wettest season, while the American ones (P. luteola) thrive in areas that are progressively warmer in this period. This suggests that these two taxa have clearly different (macro)climatic requirements, that do not depend on the different types of climate available in each continent. This supports the idea that they may be not the same species having different ecological requirements.

Keywords: Atlantic Disjunction, Biogeography, Ecological Niche Factor Analysis, Niche Overlap, Sister Taxa

Not as straightforward as expected: Disentangling the genetic structure of *Funaria hygrometrica* along a climatic gradient within Spain using genotyping-by-sequencing (GBS)

Rosa M. Ros¹, Elena de la Cruz-Martínez¹, Olaf Werner¹, Jairo Patiño² & Susana Rams³

¹Department of Plant Biology, Faculty of Biology, University of Murcia, Campus de Espinardo, 30100 Murcia, Spain (<u>rmros@um.es</u>; <u>elenala.cruz1@um.es</u>; <u>werner@um.es</u>)

²Department of Botany, Ecology and Plant Physiology, Faculty of Pharmacy, University of La Laguna, Campus Anchieta, 38200, San Cristóbal de La Laguna (Santa Cruz de Tenerife), Spain (jpatino@ull.edu.es)

³Department of Teaching of Experimental Sciences, Faculty of Education Sciences, University of Granada, Campus de La Cartuja, Granada, Spain (<u>susanarams@ugr.es</u>)

There are a number of reasons that support why bryophytes are ideal models for studies in molecular ecology and evolution. Among others, the specific mechanisms influencing their diversification and the haploid nature of the dominant phase of their life cycle greatly facilitates genetic analyses, and their small size allows us to cultivate them in reduced spaces. Some of the model bryophyte species in molecular biology have a worldwide distribution, with intercontinental dispersal events having been inferred to be more frequent than previously thought. Under these circumstances, the question arises whether these ubiquitous species exhibit genetic variants that are locally adapted to certain types of habitats, or whether there is a "generalist genotype" which supports very different climatic conditions. In the present study, a total of 183 specimens of the cosmopolitan moss Funaria hygrometrica were sampled from 80 locations spread over several biogeographical areas of Spain influenced by different climates, ranging from the Mediterranean climate in the south (The Sierra Nevada Mountains and Murcia province) to the Atlantic Ocean influenced climate in the north (Asturias and Leon provinces). The Sierra Nevada Mountains present a high altitudinal range from 0 to 3500 m. For molecular analysis all the specimens were grown in vitro under axenic conditions from individual spores. Genotyping-by-sequencing (GBS) technique was employed to obtain two multiplexed DNA libraries and identify single-nucleotide polymorphisms (SNPs). Based on the GBS data, F_{ST} values between populations were low with the exception of the high altitude populations from Sierra Nevada Mountains, which were clearly differentiated from all other populations, even the neighbouring Sierra Nevada lowland populations. These results were confirmed by additional analyses, including genetic distance, PCA and Structure-like approaches. Our data seem to exclude geographic distance as a relevant mechanism to explain the observed differences between the highland and lowland populations of *F. hygrometrica* within Sierra Nevada. Further analyses are needed in order to support the hypothesis that climatic factors are responsible for the observed distribution pattern of genetic variants or whether other factors (e.g. historical migrations, human influence) might be the main drivers.

Keywords: molecular ecology, population genomics, plant molecular genetics, cosmopolitan bryophytes

Ten years of bryological research in Croatia - Where we were, where we are, where we are going

<u>Vedran Šegota¹</u>, Antun Alegro¹, Anja Rimac¹, Beàta Papp²

¹ Division of Botany, Department of Biology, Faculty of Science, University of Zagreb, Marulićev trg 20/II, 1000 Zagreb, Croatia (vedran.segota@biol.pmf.hr, antun.alegro@biol.pmf.hr, anja.rimac@biol.pmf.hr)

² Hungarian Natural History Museum, Könyves Kálmán krt. 40, 1087 Budapest, Hungary (pappbea@bot.nhmus.hu)

Despite the fact that Croatia is bryologically one of the most explored countries of former Yugoslavia and Southeastern Europe, the data are to a large extent spatially and temporally biased. Beginning in the first decades of the 19th century, the research had the highest peak at the end of the 19th and at the beginning of the 20th century, intensified again in the 1950s and 1960s, followed by the almost complete absence of any bryological work until 2009. Thenceforward, during the last 10 years, mainly owing to cooperation between the Hungarian Natural History Museum and the University of Zagreb (Croatia), 58 new taxa were discovered as new for Croatia. Apart from this, in order to summarise all historical literature and herbarium data, a publically available database with nearly 31.000 geocoded spatio-temporal entries has been established. Concluding, at the current state of knowledge, Croatian bryoflora includes 789 taxa - 606 mosses, 178 liverworts and five hornworts. Future studies should be focused on (1) Croatian regions with lack of bryological data, (2) many taxa with single and very old records and (3) threatened species and species of conservation value.

Keywords: bryoflora, database, new taxa, Southeastern Europe,

Species richness and communities of bryophytes in heath forest, Khao Ngon Nak Mountain, Krabi Province, southern Thailand

Athita Senayai¹, Worapoj Lomlim², Sahut Chantanaorrapint¹

¹ Department of Biology, Faculty of Science, Prince of Songkla University, Hat Yai, Songkla, 90112, Thailand (athita.senayai@gmail.com); ² Hat Noppharat Thara - Mu Ko Phi Phi National Park, Ao Nang, Krabi, 81180, Thailand

Previous studies have shown that the heath forests are unique habitats and harbor an interesting bryophyte flora. Unfortunately, most of them have been disturbed by human activities, the undisturbed forests only persist as separated fragments. Consequently, the aim of this study was to determine the species richness and communities of corticolous and terrestrial bryophytes in the heath forest located at the summit of Khao Ngon Nak Mountain, Krabi Province, southern Thailand. The collection and investigation of bryophytes were carried out during February-April 2019. The corticolous bryophytes were taken from 40 selected tree trunks at 0.8 to 1.5 m height above the ground whereas the terrestrial bryophytes were collected from 40 guadrats of 2.5 square meters. The species richness, cover and life form of bryophytes were recorded during this study. In total, 43 species distributed in 27 genera and six families of bryophytes were found, which include 26 species of liverworts and 17 species of mosses. Among these, the most common families of liverworts and mosses were Lejeuneaceae and Calymperaceae, respectively. In addition, Acromastigum echinatum (Gottsche) A. Evans and Pycnolejeunea cavistipula (steph.) Mizut are reported as new species records for Thailand. According to the microhabitats, there were 37 corticolous species and 15 terrestrial species. Based on Two-way cluster analysis, four communities of corticolous bryophytes and six communities of terricolous bryophytes could be recognized. The most common life form of corticolous bryophytes was mats whereas turfs corresponded to most of the terricolous bryophytes.

Keywords: Bryophyte species richness, heath forest, bryophyte community

Observations of anisomorphic spores in Portuguese populations of *Homalothecium sericeum* (Hedw.) Schimp. *s.s.* and *H. meridionale* (M. Fleissch. & Warnst.) Hedenäs (Brachytheciaceae)

¹<u>Cecília Sérgio</u>, ²Manuela Sim-Sim, ¹César A. Garcia

¹Universidade de Lisboa, Museu Nacional de História Natural e da Ciência/CE3C-Centre for Ecology, Evolution and Environmental Changes. Rua da Escola Politécnica, 58, 1250-102 Lisboa, Portugal. (<u>csergio@fc.ul.pt</u>); ²Universidade de Lisboa, Faculdade de Ciências, Departamento de Biologia Vegetal/CE3C-Centre for Ecology, Evolution and Environmental Changes. Lisboa, Portugal.

Despite the normal homospory in bryophytes, two ranges of spore sizes (anisosporous) have been reported mostly in dioicous species (Korpelainen, 1998; Glime & Bisang, 2017) and *Homalothecium* Schimp. is a dioicous genus. Evidences have been described that in some bryophytes male and female spores are unequally sized, and that smaller spores will produce male plants (Ramsay, 1979). On other hand, some publications referred the presence of dwarf males in *Homalothecium* Schimp. (Sérgio *et al.*, 2003; Rosengren & Cronberg, 2014).

In this work, the anisosporic condition was analysed in individual capsules of selected samples of *Homalothecium sericeum* (Hedw.) Schimp. s.s. and *H. meridionale* (M. Fleissch. & Warnst.) Hedenäs, and the observed values of spore size present a distinct bimodal distribution in both species. Some spores were photographed under scanning electron microscopy to reveal the anisospory of these *Homalothecium* species using herbarium samples.

In conclusion, this study shows that we are facing a typical phenomenon of anisospory, and we can suspect that the occurrence of anisospory is linked to the presence of dwarf males in *Homalothecium sericeum s.l.*

Glime J.M. & Bisang I. 2017. Sexuality: Size and Sex Differences. Chapt. 3-3. In: GLIME J.M. Bryophyte Ecology. 3-3-1 Volume 1. Physiological Ecology. Ebook sponsored by Michigan Technological University and the International Association of Bryologists. Last updated 30 March 2019 and available at http://digitalcommons.mtu.edu/bryophyte-ecology/.

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Rosengren F. & Cronberg N. 2014. The adaptive background of nannandry: dwarf male distribution and fertilization in the moss *Homalothecium lutescens*. Biological Journal of the Linnean Society 113, 74–84.

Sérgio C., Diogo A.L. & Garcia C. 2003. O género *Homalothecium* Bruch & Schimp. em Portugal. Dados Taxonómicos e corológicos. In Notulae Bryoflorae Lusitanicae IX. 2. Portugaliae Acta Biologica 21: 282–285.

Keywords: Pleurocarpous moss, dioicous moss, anisosporic condition, scanning microscopy, dwarf males

Developmental stage determines the accumulation pattern of UV-absorbing compounds in the model liverwort Marchantia polymorpha subsp. ruderalis under controlled conditions

<u>Gonzalo Soriano^{1 2}</u>, M. Ángeles Del-Castillo¹, Laura Monforte¹, Javier Martinez-Abaigar¹ & Encarnación Núñez-Olivera¹

¹ Dpto. Agricultura y Alimentación, Universidad de La Rioja, C/ Madre de Dios, 53-26006, Logroño, España. (gonzalo.soriano@unirioja.es); ² Dpto. Genética Molecular de Plantas, Centro Nacional de Biotecnología, CSIC, Calle Darwin, 3, 28049 Madrid, España.

The liverwort Marchantia polymorpha subsp. ruderalis is emerging as a new model plant, but relatively little is known on basic aspects of its physiology and responses to environmental factors (particularly, UV radiation). Thus, our aim was to study the influence of the developmental stage on the accumulation patterns of phenolic UV-absorbing compounds (UVACs) in samples exposed and non-exposed to UV radiation. Samples of three different developmental stages (gemmae, one-month-aged thalli and two-month-aged thalli) were exposed to only PAR (P regime) and to a combination of PAR + UV-A + UV-B radiation (PAB regime) for 38 days, using moderate realistic UV doses. At the end of this period, the bulk level of methanol-soluble UVACs and the contents of six soluble individual UVACs (derivatives of apigenin and luteolin) were measured.

Both the radiation regime and the developmental stage strongly influenced UVACs accumulation pattern, and the responses to UV radiation were different in the diverse developmental stages. Plants exposed to UV from juvenile stages (gemmae or 1-month age thallus) were the most UV-responsive and showed a strong increase in every UVAC measured, being the increase stronger in the most juvenile stage. However, when samples were exposed to UV from 2-month age thalli, only apigenin derivatives increased and more modestly. In fact, the phenolic profile of these plants was not very different from the profile of the plants non-exposed to UV. Thus, as age increased, the responses to UV were weaker, probably due to senescence and a consequent decrease in metabolic responsiveness. Nevertheless, the thalli became progressively tougher due to continuously decreasing water content, representing a possible structural protection against excess UV. On the other hand, 1-month age thalli exposed to UV radiation showed a transitory conversion of apigenin mono-glucuronide into apigenin di-glucuronide. In plants non-exposed to UV, temporal variations of UVACs were much less marked and compound dependent. A PCA ordination of the different samples summarized the findings described above.

The pattern described should be taken into account in experiments in which accumulation of UVinducible compounds is expected, in order to select the adequate thalli age and better interpret the results obtained.

Keywords: Marchantia, physiologic UV responses, phenolic compounds, age-dependent.

The moss flora of Borneo: Progress and challenges

Monica Suleiman

Institute for Tropical Biology and Conservation, Universiti Malaysia Sabah, Jalan UMS, 88400 Kota Kinabalu, Sabah (email: monicas@ums.edu.my)

This paper presents the progress and challenges of completing the moss flora of an island as large as Borneo, which is politically divided by three countries, namely Malaysia (Sabah and Sarawak states), Indonesia (Kalimantan province) and Brunei. The previous catalogue of mosses from Borneo was published in 2006 which reported 721 taxa distributed in 185 genera and 50 families. Since then, 16 new localities in Sabah and one in Sarawak were studied by the author, including moss floras of large protected areas such as the Crocker Range Park, Imbak Canyon Conservation Area, Nuluhon Trusmadi Forest Reserve and Maliau Basin Conservation Areas. There were a couple of surveys in Brunei and Sarawak by other Malaysian bryologists but none in Kalimantan, which accounts for twothirds of the island's territory. Kalimantan was not explored by Malaysian bryologists due to accessibility, more costly and research permit requirement. The current number of mosses reported for Borneo is 766, an increase of 47 taxa since 2006. The highest number of species is from Sabah, followed by Sarawak and East Kalimantan. Sabah had the highest additions of new records, followed by Brunei and Sarawak, with 71, 68 and 34 taxa, respectively. The lowest was Kalimantan with only three additions. Efforts need to be given to Kalimantan though the "Heart of Borneo" initiative, a transboundary effort agreed by the three countries to enable conservation and sustainable development. The Bornean moss flora is far from being completely surveyed. More students also need to be trained to carry out bryological work in less explored areas in Borneo.

Keywords: Brunei, Kalimantan, mosses, Sabah, Sarawak

A Calcium-Dependent Protein Kinase Gene *RcCDPK* from *Racomitrium canescens* Enhanced Plant Drought and Salt Tolerance

Wei Sha, Weiwei Ren, Weiwei Zhu, Meijuan Zhang, Tianyi Ma

Heilongjiang Provincial Key Laboratory of Resistance Gene Engineering and Protection of Biodiversity in Cold Areas, College of Life Science, Agriculture and Forestry, Qiqihar University, Qiqihar, 161000, China (shw1129@263.net, tyma8902@hotmail.com)

Drought has already been a globally severe issue, crop decrease caused by drought led to great loss every year. For enhancing drought-resistance of plants, finding out functional drought-resistance related genes from plants with drought-tolerance is an effective and important way. Racomitrium canescens is a moss with great drought-tolerance and rehydration ability, a calcium-dependent protein kinase gene named RcCDPK from R. canescens was found in response to dehydration and rehydration treatments in mRNA expression level, and was selected for functional analysis in this study. Firstly, the expression of RcCDPK was furtherly determined by qPCR in R. canescens under different desiccation and rehydration treatments, the expression of RcCDPK was dramatically increased after desiccation treatment for 20 min then decreased, and also changed differently in rehydration treatments, suggesting the potential of RcCDPK participating drought resistance responses of R. canescens. For further functional verification, RcCDPK overexpression transgenic Arabidopsis thaliana strains were constructed for researches. T₃ generation RcCDPK overexpression transgenic plants showed better phenotypes under 150 mmol/L NaCl, 6% and 8% treatments. The RcCDPK overexpression transgenic Arabidopsis performed longer root length, more rosetted leaves and bigger rosette leaf size, more and longer lateral roots existed under stress treatments compared with wild type Arabidopsis. This study demonstrated overexpression of RcCDPK will increase drought and slat stress tolerance of Arabidopsis thaliana, inferring the genes from R. canescens can be used to improve properties of other plants and more stress resistance related genes from R. canescens may be valuable to be explored.

Keywords: Racomitrium canescens, RcCDPK, drought stress, salt stress

Genetic and morphological variation in the circumpolar distribution range of Sphagnum warnstorfii

Yousefi N.¹, <u>Mikulášková E.²</u>, Stenøien H. K.¹, Flatberg K. I.¹, Košuthová A.^{2,3}, Hájek M.² & Hassel K.¹

¹NTNU University Museum, Norwegian University of Science and Technology, NO-7491 Trondheim, Norway; ²Department of Botany and Zoology, Faculty of Science, Masaryk University, CZ-611 37 Brno, Czech Republic (evamikul@gmail.com); ³Department of Cryptogams, Plant Science and Biodiversity Center, Slovak Academy of Sciences, 845 23 Bratislava, Slovakia

The Quaternary climatic oscillations caused pronounced changes in the distribution of the genetic variation among populations as well as phenotypic diversification worldwide. We studied how important are these processes in plants with high gene flow potential - peat mosses (*Sphagnum* sp. div.). High level of phenotypic plasticity is common for peat mosses and it should be caused by both local habitat conditions and early cladogenesis.

Sphagnum warnstorfii is a dioicous species that occurs in intermediate and rich fens and calcareous moss tundra. Phenotypic variation has been observed among plants from the arctic zone. The previous studies show signals of genetic adaptation to microhabitat calcium and magnesium levels, however, study area was limited to the Central Europe. In this study, we aimed to explore the genetic structure of *S. warnstorfii* throughout its wide distribution. We want to investigate whether morphological divergence in arctic and boreal (boreo-nemoral) plants has a genetic basis, whether diversifying evolution has taken place and to what extent this might have resulted from vicariance events during the Quaternary period.

We used single nucleotide polymorphism genotyping to investigate the patterns of genetic diversity in 112 plants (105 localities) sampled throughout the species distribution range and explored correlation with phenotypic variation.

Genetic cluster analysis identified two main genetic lineages with an average F of 0.21 between them, and patterns of genetic structuring are highly correlated with morphological differentiation. The first cluster is restricted to the Arctic region, whereas the second has a wider distribution range covering the Arctic, boreal and boreo-nemoral regions of Eurasia and North America. The first cluster is morphologically stable also in areas where both clusters co-occur. However genetic differentiation between clusters are not high that's why we conclude that, for the time being, there is no clear evidence that the two *S. warnstorfii* morphs represent separate species.

Based on approximate Bayesian computation simulations, we find that the two lineages probably diverged from each other before the Last Glacial Maximum. It seems likely that the first cluster has survived the glacial cycles in the northernmost areas, in *'in situ'* refugia and/or in unglaciated regions of Beringia.

Yousefi N., Mikulášková E., Stenøien H. K., Flatberg K. I., Košuthová A., Hájek M. & Hassel K. (2019): Genetic and morphological variation in the circumpolar distribution range of Sphagnum warnstorfii: indications of vicariant divergence within a common peatmoss. - Botanical Journal of the Linnean Society, 189, 408-423.

Keywords: Arctic, bryophytes, cluster analysis, divergence, glacial refugia.

A Revision of Family Bryaceae sensu lato (Musci) in China*

Jiancheng ZHAO¹, Yongying LIU², Yulu NIU³, Xiaorui WANG¹

(1. Hebei Normal University, Shijiazhuang 050016, China; 2. Jiaozuo Teachers College, Jiaozuo 454001, China; 3. Hengshui University, Hengshui, 053000, China)

This paper focuses on the research classification of Bryaceae (s. l.) in China. Morphological characters of more than 4000 specimens of the Bryaceae (s. l.) and its related groups in China were analyzed. Combining morphological and molecular data in the analyses showed that: (1) Not only were the Mielichhoferia group and the family Bryaceae in the different clade in the molecular tree, but also morphological features, such as the leaf shape, the leaf cells of the two groups were different. Mielichhoferia group should be separated from the Bryaceae. And then, no morphological synapomorphy can be detected for Mielichhoferia group and the family Mniaceae, although them were in the same clade in the molecular tree. So we adopted views that the Mielichhoferiaceae was circumscribed as a single family. There were five genera, Mielichhoferia, Pohlia, Pseudopohlia, Epipterygium and Synthetodontium, in the Mielichhoferiaceae in China, total 36 species and 2 variaties. (2) The genera Orthodontium and Leptobryum were respectively transferred from Bryaceae (l. s.) to family Orthodontiaceae and Meesiaceae. (3) Members of re-circumscribe Bryaceae include 5 genera, Anomobryum, Brachymenium, Bryum, Plagiobryum and Rhodobryum in China, total 77 species and 2 variaties. Some intraspecies changes or interspecies changes have been made for Bryaceae (l. s.) also. Synthetodontium kunlunense J.C. Zhao et Y.Y. Liu and Haplodontium zangii X.R. Wang & J.C. Zhao were named as new species. Three new combination, Synthetodontium gossypinum (X.J. Li & M. Zang) J.C. Zhao & Y.L. Niu, Haplodontium himalayanum (Mitt.) X.R. Wang & J.C. Zhao and H. sinensis (Dixon) X.R. Wang & J.C. Zhao were proposed. Bryum blandum ssp handelii (Broth.) Ochi was treated as B. handelii Broth. B. petelotii Thér. & Henr. and B. yuennanense Broth. were treated as independent species. Six species of Bryaceae (l.s.) were reported for the first time from China, e.g., Bryum amblyodon Müll. Hal., B. weigelii Spreng, B. moravicum Podp., Pohlia andrewsii A.J. Shaw, P. andalusica (Höhn.) Broth. P. marchica Osterw. and Anomobryum concinnatum (Spruce) Lindb. were resurrected for species. Pohlia minor Schleicher ex Scheaegr. was reduced as P. elongata var. greenii (Brid.) Shaw. Three propaguliferous species, Pohlia flexuosa Harv., P. leucostoma M. Fleisch. and P. proligera (Lindb. ex Breidl.) Lindb. ex Arn. were correct identified in China.

*This work was supported by the National Natural Science Foundation of China (Grant Number 31370237).

Key words: classification, Mielichhoferiaceae, molecular tree, morphological character, sensu lato Bryaceae

Presentation type: Poster Presentation

Flora of Bryophytes and their Ecological Distribution in Karst Tiankeng, Southern China

Zhang Zhaohui¹, Wang Zhihui², Wang Dengfu¹, Wu Qimei¹

¹ Key Laboratory for Information System of Mountainous Area and Protection of Ecological Environment of Guizhou Province, Guizhou Normal University, Guiyang 550001, China(zhaozhang9@hotmail.com).² School of Life Sciences, Guizhou Normal University, Guiyang Guizhou 550001, China;

Karst Tiankengs (limestone sinkholes or collapse dolines) are special landforms with unique bryophyte flora. Some huge and interesting Tiankengs were found in recent three decades from China, such as Xiaozhai Tiankeng (the depth 662m, length 625m, width 535m,) and Haolong Tiankeng (the depth 520m, length 809m, width 426m,). This work aims to carry out some preliminary study on flora of bryophytes and their ecological distribution from karst tiankengs. From July 2017 to March 2019, supported by the National Natural Science Foundation of China (NSFC. No.31760050, No.31760043, No.31360043 and No. 31360035), many surveys were conducted in southern China, about 2640 samples and specimens were collected from 18 karst Tiankengs.

As main results, 236 species distributed in 78 genera and 24 families were found in Karst Tiankeng areas in Guizhou, Chongqing and Guangxi province, China. The phytogeographical elements of the bryoflora include: Tropical Asian (18.64%), Tropic Asian to Tropic African (2.12%), Pantropical (3.39%), North Temperate (20.34%), East Asian (20.33%), East Asian and North America disjuncted (4.24%), Old World Temperate (2.12%), Temperate Asian elements (3.38%), Endemics to China(12.71%) and Cosmopolitans (13.56%). Ten types of bryophyte life-forms were found distribution in Karst Tiankengs. Among them, Tall turfs, Mats, Wefs and Dendroids were dominant , which suggested that the whole sinkhole is dark, damp and shadow. The vertical distribution of bryophytes caused by gradient effect of the light, humidity and temperature, and the diversity of bryophyte communities was the result of the combinations of three environmental factors and human activity. The rich bryophyte flora indicates that Karst Tiankeng is a natural refuge for bryophyte conservation in the vast karst area from southern China.

Keywords: Bryophytes; flora; ecological distribution; karst Tiankeng (Sinkhole), China

Presentation type: Poster Presentation

Diversity of moss peristomes and their distribution in karst fengcongs(Cone Karst) in Guizhou Plateau, China

Wang Zhihui¹, Zhang Zhaohui²

¹ School of Life Sciences, Guizhou Normal University, Guiyang Guizhou 550001, China (<u>wangzhihui222@126.com</u>); ² Key Laboratory for Information System of Mountainous Area and Protection of Ecological Environment of Guizhou Province, Guizhou Normal University, Guiyang 550001, China(<u>zhaozhang9@hotmail.com</u>).

Peristome is an important structure for mosses, but related research on this issue is very weak in karst plateau from both China and abroad.

From October 2017 to January 2019, supported by the National Natural Science Foundation of China (NSFC. (No.31760043, No.31760050), we carried out bryological surveys in the three typical karst fengcongs(Cone Karst) of Guizhou, China, in the course of which about 860 samples were collected. A hundred-eight taxa of mosses were identified from these samples.

Among them, 42 taxa were studied focusing on the diversity of moss peristome structure by means of Optical microscope and Electron Microscope, and the SDR, Sorenson similarity index and RDA were used for analysis of the ecological distribution and the effect of environmental factors. As main result, two moss groups of peristome morphology were found: one group is an Arthrodontae peristome with 38 taxa, the other group is a Nematodontae peristome with only 4 taxa. Among the Arthrodontae mosses, there were 16 taxa belonging to Acrocarpi-haplolepideae, 10 species in Acrocarpi-diplolepideae and 12 taxa in Pleurocarpi-diplolepideae, respectively. The results showed that: the Pleurocarpi-diplolepideae group was mainly distributed at the bottom of the karst fengcong, while the Acrocarpi-haplolepideae mosses dominated the middle and top of the fengcong. The RDA analysis showed that the distribution of the Acrocarpi-haplolepideae mosses was mainly influenced by the light intensity and the environmental temperature; The the distribution of the Peurocarpi-diplolepideae mosses was mainly influenced by the relative humidity; and the distribution of the Acrocarpi-diplolepideae mosses was mainly affected by the altitude. From the top to the bottom of the karst fengcong, the distribution of the Arthrodontae mosses also changed regularly with the gradient decreasing of the relative humidity. The results provide some interesting data to study the diversity of moss peristomes and conservation mosses in karst mountain areas in southwest China

Keywords: Moss, peristomes, ecological distribution, karst fengcongs, -Plateau

The genus Mitthyridium (Calymperaceae, Bryophyta) in Thailand

Santiporn Boonjing¹, Orawanya Suwanmala¹, Si He², & Sahut Chantanaorrapint¹

¹Department of Biology, Faculty of Science, Prince of Songkla University, Hat Yai, Songkhla 90112, Thailand (Santipornb@gmail.com); ²Missouri Botanical Garden, 4344 Shaw Blvd., St. Louis, MO 63110, U.S.A.

The genus *Mitthyridium* H.Rob. in Thailand is revised. The study was based on a morphological study of fresh specimens collected throughout the country as well as herbarium specimens housed in BKF, BM, EGR, FH, MO, NY, and PSU. Eight species and three subspecies of the genus are recognized: *M. constrictum* (Sull.) H.Rob., *M. fasciculatum* (Hook. & Grev.) H.Rob. subsp. *cardotii* (M.Fleisch.) A.Eddy, *M. fasciculatum* subsp. *fasciculatum*, *M. fasciculatum* subsp. *obtusifolium* (Lindb.) M.Menzel, *M. flavum* (Müll. Hal.) H. Rob., *M. jungquilianum* (Mitt.) H.Rob., *M. luteum* (Mitt.) H.Rob., *M. repens* (Harv.) H. Rob., *M. undulatum* (Dozy & Molk.) H.Rob., and *M. wallisii* (Müll. Hal.) H.Rob. Of these, three species and one subspecies are newly recorded in the country: *M. fasciculatum* subsp. *obtusifolium*, *M. luteum*, *M. undulatum*, and *M. wallisii*. *Mitthyridium* is widely distributed in the southern half of the country; it does not seem to occur in the northern part. The greatest diversity of species was found in the peninsular region. The most common species of *Mitthyridium* in Thailand is *M. flavum* while *M. constrictum* was found only in Phangnga province.

Keyword: Bryophyta, Calymperaceae, Mitthyridium, mosses, Thailand

DIVERSITY, DISTRIBUTION AND SOCIO-CULTURAL IMPORTANCE OF BRYOPHYTES IN THE SUDANO-GUINEAN AND SUDANIAN AREAS IN BENIN

<u>Alia M. H. Boris¹</u>, Reeb Catherine², Tossou G. Monique¹, Akoegninou Akpovi¹ 1-Department of Plant Biology, Laboratory of Botany and Plant Ecology, University of Abomey-Calavi, Benin

2-Laboratory of the Institute of Systematic Evolution of Biodiversity (ISYEB) in Paris, University of Sorbone in Paris, France. catherine.reeb@gmail.com/catherine.reeb@mnhn.fr

Benin exhibits very few bryological studies. These were conducted by foreign authors in a part of the country, leading to a great inadequacy and insufficient knowledge of the bryophyte flora of Benin. Our work was carried out in the Sudano-Guinean and Sudanian areas of Benin. We then sought to: (1) Inventory and set the distribution of bryophyte species; (2) Determine the ecological and anthropogenic factors explaining their distribution; (3) Document traditional knowledge related to bryophytes and determine the socio-cultural and ecological factors influencing this traditional knowledge. Bryophyte distribution models were examined in three microhabitats in closed (forest) and open (savannah) environments. The uniform nature of the environments provided an excellent system for studying the relationship between environments and species, diversity and distribution, while avoiding confusing the multiple effects of substrate and vegetation heterogeneity. The bryophytes were studied using 50 X 50 m blocks on the ground, on the hills and in the waterfalls. A semi-structure interview survey of traditional therapists was conducted to collect local names, uses, treated conditions, preparation methods and different forms of use of bryophytes. In our study, the diversity of bryophytes increases with the closed environment. The species richness is very high; sixty bryophyte species have been identified. Diversity was also controlled by ecological conditions of the microhabitats (humidity, shade). Tillage and soil moisture seem to exert a strong selection on species capable of living in a given environment. These factors are therefore relevant to explain the distribution of specific diversity as well as the dynamics of bryophyte population in Benin. The results reveal that the diversity of Benin bryophytes is partly favoured by the richness in mineral substrate (limestone). Twenty-five species have been related to the treatment of forty-two diseases. They are involved in the preparation of sixty recipes. Decoction and maceration are the most frequent methods of preparation. Educational level, ethnicity and ecological zone determine ethnobotanical knowledge. Mineral environments and the rest of Benin tropical forests should be preserved for their high bryophyte diversity.

Keywords: Bryophytes, microhabitat, Sudano-Guiana, Sudanese, Tradiotherapists.

A contribution to the knowledge of the bryophytes of the Cape Verde Islands, with an emphasis on Santo Antão and São Vicente

<u>César Augusto Garcia¹</u>, Cecília Sérgio^{1,2}, Anabela Martins¹, Ana S.B. Rodrigues², Manuela Sim-Sim^{1,2}

¹ Universidade de Lisboa. Museu Nacional de História Natural e da Ciência/Centre for Ecology, Evolution and Environmental Changes. Natural History and Systematics (NHS) Research Group (cE3c). Rua da Escola Politécnica, 56/58. 1250-102 Lisboa, Portugal.

² Universidade de Lisboa. Faculdade de Ciências. DBV/cE3c 1749-016, Lisboa, Portugal

During 2016 Santo Antão and São Vicente in the Cape Verde archipelago were visited and surveyed for bryophytes. Five species are reported new to Cape Verde, *Cheilolejeunea rigidula* (Nees ex Mont.) R.M.Schust, *Riccia trabutiana* Steph., *Lindbergia patentifolia* Dixon, *Lewinskya acuminata* (H.Philib.) F.Lara, Garilleti & Goffinet and *Timmiella cameruniae* Broth. The presence of *Porella canariensis* (F.Weber) Underw in the archipelago is also confirmed. Additionally, *Frullania spongiosa* Steph., *Bryum dichotomum* Hedw., *Cryptoleptodon longisetus* (Mont.) Enroth and *Didymodon hastatus* (Mitt.) R.H.Zander are reported for the first time from Santo Antão island. *Cheilolejeunea rigidula*, *Lindbergia patentifolia* and *Timmiella cameruniae* are new to Macaronesia.

Keywords: Biodiversity, Bryophyte flora, Africa, Macaronesia, Atlantic islands

Which bryophytes are preferred by bryophagous insects?

Michaela Drgová¹, Petr Pyszko¹, Vítězslav Plášek¹, Pavel Drozd¹

¹Department of Biology and Ekology, University of Ostrava, Chittussiho 10, 710 00 Ostrava, Czech Republic (<u>misadrgova@gmail.com</u>)

Bryophytes provide an ideal environment for great diversity of arthropods and also a food source for remarkable guild of invertebrate bryophage. Since proper and unbiased estimates of bryophages density cannot be made by any standard sampling method (including sieving) but only by thorough examination of bryophyte cushions, the host preferences of bryophages have been almost exclusively concluded from observations of individuals on particular host mosses. We focused on largest group of insect bryophages – Byrrhidae (Coleoptera) to find their preferences for bryophytes. We sampled 416 quadrates (0.25 m² each quadrate) of 48 species of bryophytes across 26 localities and we examined them thoroughly for presence of bryophagous and bryobiontic invertebrates. Additionally, we kept and repeatedly checked the samples to raise insect juvenile stages. We analysed data using generalized linear mixed models and multi-level pattern analysis to find: a) differences in beetle density in different moss taxa, b) the associations between particular byrrhid species and bryophytes. The density of Byrrhidae differs strongly among moss species and this pattern could be simplified on moss orders. Filtering only mosses with sampled area $> 2.5 \text{ m}^2$, the greatest densities of beetles we found on Pohlia nutans, Polytrichum piliferum, Dicranella heteromalla, Barbula unguiculata, Bryum argenteum, Brachythecium rutabulum or Ceratodon purpureus, whereas very low or zero densities were on Hypnum cupressiforme, Dicranodontium denudatum, Thuidium tamariscinum, Syntrichia ruralis or Didymodon rigidulus. We did not find significant differences between host preferences for observed byrrhid species. Our results bring first insights in host preferences of bryophages based on intensive quantitative sampling.

Keywords: bryophagous beetles, Byrrhidae, host preferences, host specificity.

Information for submission of abstracts to the Bryology 2019 meeting. Deadline: 31 May 2019

Presentation type: Oral Presentation, Poster Presentation

Abstract Title: The Phenotypic Plasticity of *Philonotis hastata* (Duby) Wijk & Margad.

Mohamed Farag^{1,2}, Hanna Shabbara¹, Wagieh El-Sadaawi¹

1 Botany Department, Faculty of Science, Ain Shams University, 11566 Egypt

2 Botany Department, Faculty of Biology, Murcia University, 30100, Spain

e-mail of presenting and corresponding author: <u>mohamed_farag@sci.asu.edu.eg</u>

Bryophytes are undergoing many morphological variations, physiological adaptations and various reproductive strategies since they colonized terrestrial habitats. *Philonotis* Brid. (Bartramiaceae) has a high level of identification difficulty because of the existing intergradation between its species. This study aimed to assess the phenotypic plasticity of *P. hastata* (Duby) Wijk & Margad. under different strength of Murashige and Skoog "MS" nutrient medium. Approximately 40 characters of protonema and gametophyte stages were recorded. The shape of gametophytes were more or less similar in half strength MS and full strength MS to typical form of *P. hastata* in natural habitats. While in zero strength MS medium caused phenotypic changes of the gametophyte of *P. hastata*. It had triangular leaf lamina with excurrent costa and slightly revolute margin, and it lost the ability to form detachable branches. These overall characters are typical of either *P. falcata* (Hook.) Mitt., *P. marchica* (Hedw.) Brid. or *P. rigida* Brid.. The phenotypic plasticity of *P. hastata* opens a new dimension towards the study of the speciation in *Philonotis*.

Keywords: *in-vitro* cultivation, morphological variation, *Philonotis*, speciation.

Distribution pattern of epiphytic mosses from Orthotrichaceae family in western provincies of China

Lucie Fialová¹, Lukáš Číhal^{1, 2}, Zuzana Komínková¹, Vítězslav Plášek¹

¹ Department of Biology and Ecology, University of Ostrava, Chittussiho 10, 710 00 Ostrava, Czech Republic (fialova.lucie92@gmail.com); ² Silesian Museum, Research and Collections; Nádražní okruh 31, 746 01 Opava, Czech Republic (cihal@szm.cz)

Species distribution modeling (SDM) is very widespread method how to find out species distribution pattern of species. Despite the frequent used of this method there are still few studies how to evaluate the potential current distribution structures and spread of epiphytic mosses from Orthotrichaceae family. The main aim of this research is to find out which kind of environmental conditions and how way they can influence the distribution structure of selected species from Orthotrichaceae family in western provinces (Sichuan, Yunnan, Xizang, Gansu, Xinjiang a Qinghai) of China. For our purpose we have chosen the following five epiphytic mosses for SDM: Lewinskya hookeri, Nyholmiella obtusifolia, Orthotrichum alpestre, O. pamiricum, and O. scanicum. Named species are rare and endangered in the world scale. The obtained results can contribute to the conservation and get better understanding to ecological requirements of target species. As the basic data for the evaluating of species distribution pattern of selected epiphytic mosses, we used data from Chinese herbarium collections and field survey. A total of 203 samples (presence-only data) were analysed. The obtained data were evaluated by using Maxent software, R and Qgis for data preparation and individual environmental layers. Maxent from input data - GPS coordinates and environmental variables which we used (17 bioclimatic layers WorldClim, layer 0.5 km MODIS-based Global Land Cover Climatology, Vegetation layer (Percent Tree Cover) and slope) generates a model that predicts the most appropriate locations for the occurrence of the monitored moss species. Respectively, the model identifies regions with environmental factors that will allow the species to maintain a viable population. The final models have showed that the distribution of selected mosses is most affected by the environmental layer called LandCover. LandCover is physical material on the surface of the earth like grass, asphalt, trees, bare ground, water, etc. The model also confirmed that temperature and precipitation play an important role in the distribution of studied epiphytic mosses.

Keywords: Orthotrichum, species distribution modeling, MaxEnt

Singularity and distribution of epiphytic bryophyte communities in the NW Iberian Peninsula

Nagore G. Medina^{1,2,3}, Francisco Lara^{1,2}, Joaquín Calatayud^{3,4,5}, Isabel Draper¹, Vicente Mazimpaka¹ & Joaquín Hortal^{2,6}

¹ Departmento de Biología (Botánica). Universidad Autónoma de Madrid. C/Darwin 2, E-28049. Madrid (nagore.garcia@uam.es); ² Centro de Investigación en Biodiversidad y Cambio Global (CIBC-UAM). Departamento de Biología, Fac. Ciencias Universidad Autónoma de Madrid; ³ Departamento de Biogeografía y Cambio Global, Museo Nacional de Ciencias Naturales (MNCN-CSIC), C/José Gutiérrez Abascal 2, E-28006 Madrid, Spain; ⁴ Departamento de Ciencias de la Vida, Universidad de Alcalá, Edificio de Ciencias, Ctra. Madrid-Barcelona km. 33,6, E-28871 Alcalá de Henares, Madrid, Spain; ⁵ Integrated Science Lab, Department of Physics, Ume\r{a} University, Sweden; ⁶ cE3c – Centre for Ecology, Evolution and Environmental Changes, Faculdade de Ciências da Universidade de Lisboa, Campo Grande, 1749-016 Lisboa, Portugal

Bryophytes and vascular plants differ in many biological traits, but at the same time they occupy the same landscape and thus are subject to the influence of similar large-scale factors, such as environmental change. We search out the similarities in diversity patterns between vascular plants and bryophytes. To do so, we analyse the changes in community composition of the epiphytic bryophyte communities in the NW quadrant of the Iberian Peninsula, and compare them to the bioregions based on vascular plants. We used network modularity analysis to identify chorotypes (groups of species with overlapping range), finding four distinctive modules. Chorotype 1 was located mostly in the extreme northwestern area under Atlantic climate, and was the most singular, made out of exclusive species that were very frequent within this module but very rare outside (Hypnum andoi, Ulota crispula and U. bruchii). Chorotype 2 mostly occurred in Mediterranean mountain areas, and it was characterized by species that were more frequent inside the module but could also be found in other areas (i.e. Lewinskya acuminata, Lewinskya affinis, Hypnum cupressiforme and Homalothecium sericeum). Chorotype 3 was mainly distributed in Mediterranean plains and basins and was characterized for having species that were frequent within the module and more or less rare outside, such as Fabronia pussilla or Zygodon catarinoi. Chorotype 4, showed no clear geographic pattern and was characterized by a handful of facultative epiphytes that may be indicating anthropic influence. These modules reflect the relatively steep transition between the Atlantic and Mediterranean regions, as well as the gradual replacements between different climatic zones within the Mediterranean. Interestingly, our results have important similarities with Bolós' (1985) phytogeographic division for vascular plants. Although the precise limits of the regions are difficult to establish, the consistency in their definition across biotic groups suggests that the factors that differentiate the Atlantic and Mediterranean floras have similar effects on vascular plants and bryophytes.

Keywords: Bryophytes, community composition, chorotypes, Mediterranean region, bioregions

Taxonomic studies on type material of the family Trichocoleaceae from Central and South America with two new combinations of the genus *Leiomitra*

Tomoyuki Katagiri

Hattori Botanical Laboratory, Obi 6-1-26, Nichinan City, Miyazaki 889 2535, Japan (tomoyuki-katagiri@hattorilab.org)

The family Trichocoleaceae is one of the major components of the leafy liverworts, and over 70 species have been described in the three genera (Eotrichocolea R.M.Schust., Leiomitra Lindb., Trichocolea Dumort.). In the series of our taxonomic studies of the family, the generic concept for each genus was outlined and worldwide keys to the species of the genus Trichocolea, including eleven species, and Eotrichocolea, including two species were given. However, for the genus Leiomitra taxonomic studies were limited to Southeast Asian species. Leiomitra, which includes about fifteen species, is distributed mainly from tropical and subtropical areas of Central and South America. *Leiomitra* is distinguished from the other genera of the family by such characters as succubous leaf insertion, sublongitudinally inserted leaves, anisophyllous leaves and underleaves, and presence of a well-developed calyptra and perianth. In Central and South America, the taxonomy of the Trichocoleaceae has not been revised since Hatcher (1957) and Fulford (1963) and both authors recognized twelve species. Among these species, several remain morphologically illdefined or otherwise little-known. The taxonomic position of some of these species also remains tentative and needs critical taxonomic reassessment, since no reproductive organs or annex structures are known. In the present study, three poorly known species: Trichocolea brevifissa Steph., T. sprucei Steph., T. floccosa Herzog & Hatcher with similar morphological characters, and one common species widely distributed over Central and South America, Leiomitra flaccida Spruce, are reevaluated based mainly on their type specimens. On the basis of the presence of perianths T. brevifissa and T. sprucei are newly included in the genus Leiomitra: L. brevifissa (Steph.) T.Katag. and L. sprucei (Steph.) T.Katag. T. floccosa from Costa Rica is newly synonymized with L. sprucei. The worldwide key to the species of *Leiomitra* is given for the first time.

Keywords: leafy liverwort, Marchantiophyta, morphology, taxonomy, Trichocoleaceae

Bioactivity and phytochemical characterization of *Anoectangium thomsonii* Mitt. for the antimicrobial potential

Kavita Negi¹, Preeti Chaturvedi²

¹Central Council for Research for Unani Medicine (Ministry of Ayush), OPP-D-Block, Janakpuri, New Delhi-110 058 INDIA (<u>negikavita123@gmail.com</u>); ²Department of Biological Sciences, C.B.S.H., G.B. Pant University of Agriculture & Technology, Pantnagar 263145, Uttarakhand, INDIA.

The use of antibiotics is one of the most common means of treatment of diseases. However, prolonged use of drugs slowly develops resistance in microbial pathogens. The use of higher plants in the medicine reported from time immemorial but the lower group of plants like bryophytes is less explored. In the present study, the antimicrobial potential of ethanol and acetone extracts of *Anoectangium thomsonii* Mitt. (Moss) were studied against different microorganisms like *Staphylococcus aureus, Erwinia chrysanthemi, Bacillus cereus, Salmonella enterica, Eshcherichia coli* and *Aspergillus parasicticus*. Disc diffusion assay was done for the screening of the potent extracts and concentration dependent (100 - 1000 μ g/mL) growth inhibition was observed for all the organic extracts of Bryophytes. The minimum inhibitory concentration (MIC) and minimum bactericidal or fungicidal (MBC/MFC) was observed in employing micro dilution methods which ranged from 0.97 to 125 μ g/mL against different microbes. Among studied microorganism, *S. enterica* was found the most sensitive with lowest MIC (0.97 μ g/mL). The highest zone of inhibition (ZI=31±0.57 mm) was shown by ethanol extract of *A. thomsonii* (Dwarahat) with MIC=0.97 μ g/mL and MBC=1.95 μ g/mL. The GC-MS data obtained in ethanol extract of *A. thomsonii* showed a high percentage of sesquiterpenes, steroids, fatty acids, and alcohol derivatives attributing to its antibacterial potential.

The present study establishes the antibiotic potential of *A. thomsonii* against *S. enterica*. Ethanol extract of *A. thomsonii* possess significant potential to inhibit the growth of *S. enterica* due to presence of large number of unique chemical compounds. It will eventually help in reassuring the validation of *A. thomsonii* an ethnomedicinally important Bryophyte as a potent cure for *S. entercia* infections.

Key words: Medicinal Plants, A. thomsonii, antibacterial activity, Salmonella enterica, GC-MS.

Unravelling the Genetic Basis of the Hornwort-Cyanobacteria Symbiosis

<u>Anna Neubauer¹</u>, Manuel Waller¹, Eftychios Frangedakis², Juan Carlos Villarreal³, Fay-Wei Li⁴, Peter Szovenyi¹

1 Department of Evolutionary and Systematic Botany, University of Zurich, Switzerland (anna.neubauer@uzh.ch, peter.szoevenyi@uzh.ch); 2 Department of Plant Sciences, University of Cambridge, United Kingdom; 3 Department of Biology, Laval University, Canada; 4 Boyce Thompson Institute for Plant Research, Cornell University, USA

Plant-microbial associations are becoming increasingly recognized as major drivers of plant evolution and ecosystem dynamics. One of the most significant events in earth's history, the colonization of the terrestrial environment by the green lineage, is also postulated to have only been possible by the association with fungal and putatively with cyanobacterial partners. While functional significance of the association with fungal partners is actively investigated, plant-cyanobacterial associations of early land plants are poorly understood. Our project is meant to fill this gap and investigates the molecular and genomic bases of the plant-cyanobacterial association in the extant representatives of the earliest land plants, the hornworts.

To begin to understand the molecular bases of the plant-cyanobacteria association in details we are investigating (1) gene expression changes (RNA-seq) using a time series analysis both for the plant and the cyanobacterial partner. (2) We analyze this data using custom network analysis to identify conserved and divergent regulatory networks involved with the establishment of the association in the hornwort and in other land plants. (3) To better characterize the genes necessary for symbiosis establishment and maintenance we are currently conducting a forward genetic experiment. (4) Finally, using results of the previous analyses we will identify candidate genes and begin to verify their putative function using reverse genetics.

Our phylogenetic analysis suggests that members of the common symbiosis pathway are present in the hornwort genome. Nevertheless, a first pass analysis of gene expression dynamic during symbiosis establishment suggests that most genes of the common symbiosis pathway are not differentially expressed. This either means that communication in the hornwort-cyanobacteria symbiosis does not involve the common symbiosis pathway or genes are post transcriptionally regulated. We are currently conducting experiments to clarify this.

This study will provide fundamental information on the regulatory networks governing the plantcyanobacteria association in basal groups of land plants. Moreover, it will provide information on the conservation of gene regulatory networks involved with plant-microbial interactions in general over many million years of plant evolution. Our poster will provide information on the current state of the project.

Keywords: N-fixation, *Anthoceros agrestis*, Hornwort-Symbiosis, Cyanobacteria-Symbiosis, Forward Genetics

Bryophyte acclimation to sun conditions is influenced by both photosynthetic and ultraviolet radiations

<u>Encarnación Núñez-Olivera</u>¹, Gonzalo Soriano¹, María-Ángeles Del-Castillo-Alonso¹, Laura Monforte¹, Javier Martínez-Abaigar¹

¹ Facultad de Ciencia y Tecnología, Universidad de La Rioja, 26005 Logroño (La Rioja), Spain (encarnacion.nunez@unirioja.es)

We studied the acclimation of bryophytes to sun and shade under ambient conditions, measuring variables usually influenced by photosynthetically active (PAR) and ultraviolet (UV) radiations. Our aim was to elucidate to what extent the responses to changing radiation were influenced by PAR and UV wavelengths. For this aim, we used three taxonomically and structurally different species: the thalloid liverwort Marchantia polymorpha subsp. polymorpha, the leafy liverwort Jungermannia exsertifolia subsp. cordifolia and the moss Fontinalis antipyretica. In the field, liverworts were more radiation-responsive than the moss, and the thalloid liverwort was more responsive than the leafy liverwort. Sun plants of Marchantia polymorpha showed, in comparison to shade plants, higher sclerophylly, lower Chl a+b contents, higher Chl a/b ratios, higher (antheraxanthin + zeaxanthin) / (violaxanthin + antheraxanthin + zeaxanthin) ratios (xanthophyll index), lower F_v/F_m values, higher contents of methanol-soluble vacuolar UV-absorbing compounds (soluble UVACs), higher values of the ratio between the contents of methanol-insoluble cell wall-bound UVACs (insoluble UVACs) and soluble UVACs, higher contents of soluble luteolin and apigenin derivatives and riccionidin A, and higher contents of insoluble p-coumaric and ferulic acids. Overall, these responses reduced light absorption, alleviated overexcitation, increased photoprotection through non-photochemical energy dissipation, increased UV protection through UV screening and antioxidant capacity, and denoted photoinhibition. Jungermannia exsertifolia showed moderate differences between sun and shade plants, while responses of F. antipyretica were rather diffuse. The increase in the xanthophyll index was the most consistent response to sun conditions, occurring in the three species studied. The responses of soluble UVACs were generally more clear than those of insoluble UVACs, probably because insoluble UVACs are relatively immobilized in the cell wall. These modalities of radiation acclimation were reliably summarized by Principal Components Analysis. Using the most radiationresponsive species in the field (M. polymorpha), we found, under close-to-ambient greenhouse conditions, that sclerophylly and Chl a+b content were only influenced by PAR, F_v/F_m and luteolin and apigenin derivatives were only determined by UV, and xanthophyll index was influenced by both radiation types. Thus, responses of bryophytes to radiation can be better interpreted considering the influence of both PAR and UV radiation.

Keywords: Radiation acclimation, ultraviolet radiation, chlorophyll fluorescence, photosynthetic pigments, flavonoids

Abstract Title: Type III Phosphatidylinositol 4-kinase genes in *Physcomitrella patens:* CRISPR/Cas9 mutagenesis uncovers both novel and conserved functions in development

<u>Kaye Peterman</u>, Zijing Li, Hannah Mummey, Tatyana Johnson, Suhaily Penix, Amelia R. Cecere, Scarlett Se Yun Cheon, Linnea N. B. Fischer, Justine Hsu, Camilla Koczara, Ying Li, Emma M. Magee, Andrea Marie Sama, Mayla C. Thompson, Asriel D. Walker, Ciara Wardlow, Catherine R. Xie

Wellesley College, Wellesley, MA 02481, U.S.A. (kpeterman@wellesley.edu)

Phosphoinositides (PIs) are key signaling lipids involved in many fundamental cellular processes. In Arabidopsis, the PI, phosphatidylinositol 4-phosphate (PtdIns4P), is involved in polarized growth of root hairs and pollen tubes, chloroplast movements and division, modulation of stomatal opening/closing and salicylic acid-dependent immunity. In contrast, there is limited work on the role of PtdIns4P in bryophytes. To dissect the function of PtdIns4P in *Physcomitrella patens*, we are using CRISPR/Cas9 mutagenesis to knockout the Type III Phosphatidylinositol 4-kinases (PI4Ks) that produce Ptdins4P – PpPI4K α 1, PpPI4K α 2, PpPI4K β 1 and PpPI4K β 2. Multiplex CRISPR/Cas9 editing of the PpPI4K α 1 and PpPI4K α 2 genes yielded only single null mutants, suggesting that *pi4k\alpha1/pi4k\alpha2* double mutants are lethal. Polarized cell growth was unaffected in $pi4k\alpha 1$ and $pi4k\alpha 2$ mutants; neither the size and solidity of colonies regenerated from protoplasts, nor the size of plants grown from protonemal cell fragments, were significantly different from wild-type. However, the number of leafy gametophores produced by $pi4k\alpha 1$ was 50% of that observed on $pi4k\alpha 2$ or wild-type plants. Furthermore, the $pi4k\alpha 1$ gametophores were abnormal in morphology with irregularly shaped phyllids, composed of disorganized, misshaped cells. As PI4K α 1 is involved in the regulation of chloroplast division in Arabidopsis and in rhizoid development in Marchantia, these results point to a novel role for *Physcomitrella* PI4Ka1 in 3-dimensional growth and phyllid development. CRISPR/Cas9 mutagenesis of the PpPI4K β 1 and PpPI4K β 2 genes resulted in both single and double null mutants. Mutant $pi4k\beta1$, $pi4k\beta2$ and $pi4k\beta1/pi4k\beta2$ plants, regenerated from protoplasts, were not significantly different in size from wild-type after one-week on PPNH₄ medium. However, after 2.5 weeks, the size of $pi4k\beta 1/pi4k\beta 2$ plants was <40% that of wild-type or single mutants, suggesting an effect on polar growth of caulonemata. Growth of $pi4k\beta 1/pi4k\beta 2$, compared to wild-type, was also reduced in plants grown on PpNO₃ or in the dark, both conditions that elicit polar growth of caulonemata. These results suggest that PpPI4K β 1 and PpPI4K β 2 play overlapping roles in polar cell growth, similar to the role of these genes in flowering plants.

Keywords: phosphatidylinositol 4-phosphate, Type III Phosphatidylinositol 4-kinases, polar cell growth, phosphoinositide signalling, *Physcomitrella* development

Deciphering the role of FHY1 protein in mediating light-hormone crosstalk in *Physcomitrella patens* Durga Prasad Biswal¹, Kishore CS Panigrahi

School of Biological Sciences, National Institute of Science Education and Research (NISER), HBNI, Bhubaneswar, Jatni, Odisha, India, 752050

¹ <u>dpbiswal@niser.ac.in</u>

Phytochromes are proteins that sense red and far-red light (FR) in the environment and modulate internal responses by altering gene expression in plants. In higher plants, such as Arabidopsis thaliana, five types of phytochromes (PHYs) are present, which are categorized into type-I, PHYA (for FR) and type-II, PHYB-PHYE (for red light). Type-II phytochromes upon activation by light translocate to nucleus independently, but type-I phytochromes i.e. PHYA requires FHY1 (Far red elongated hypocotyl) protein for its nuclear translocation. This response in conserved in basal land plants such as mosses. phyA and fhy1 mutants of Arabidopsis display elongated hypocotyl as compared to wild type (WT) under FR light. However, in mosses such as *Physcomitrella patens*, *fhy1* plants do not display etiolated growth of gametophore. Rather, etiolated growth is observed in WT and *phy1* plants under FR and they show elongated gametophore. This FR light dependent growth pattern in *Physcomitrella* differs from that observed in Arabidopsis. Gametophore formation and elongation related responses are primarily controlled by hormones such as auxin and cytokinin in *Physcomitrella*. Light and hormone interaction in regulating development of Physcomitrella is poorly understood. In this study, we have investigated whether FHY1 protein plays any role in development of *Physcomitrella* under FR light in response to different phyto-hormones. To find the role of FHY1 in hormone signaling, we have evaluated protonema differentiation and bud induction pattern in WT, *fhy1* and different phytochrome mutants (namely *phy1* and phy3) under different light conditions in response to auxin and cytokinin. We observed that; (i) FR light induced caulonema formation irrespective of presence or absence of exogenous auxin in WT plants, (ii) red light promoted bud induction in WT plants in response to cytokinin in glucose dependent manner, (iii) *fhy1* plants showed defective growth under FR light as well as altered hormone sensitivity to both auxin and cytokinin. While the function of different phytochromes may be redundant for the observed responses, FHY1 appears to play major role in *Physcomitrella* development under FR light.

Key Words: Physcomitrella, Phytochrome, Phytohormone, FHY1, FR Light

Poster Presentation

The MADBRYO project, laying the groundwork and some preliminary results <u>Catherine Reeb</u>¹, Elodie Dubuisson¹, John C. Brinda²

¹ Institut de Systématique Ecologie Evolution, Sorbonne Université, MNHN, Paris, FR, (<u>catherine.reeb@mnhn.fr</u>)

² Missouri Botanical Garden, Saint Louis, US

The MADBRYO project aims to close the gaps in our knowledge <u>of</u> Malagasy bryophytes by <u>gathering</u> togethergathering the specimens and taxonomic data that are currently scattered across multiple institutions, supplementing them as necessary with targeted collecting efforts, and using them to develop a comprehensive flora for the bryophytes of Madagascar. Together, these efforts will provide a strong foundation for pursuing further scientific research on Malagasy bryophytes and for promoting their conservation.

Many historical specimens of Malagasy bryophytes have <u>been</u> accumulated in herbaria over the years. Together they represent a large and mostly untapped source of vital taxonomic and biogeographic data, but many of them have not been fully curated and are consequently difficult to use. In turn, the inaccessibility of the information they contain hampers scientific research into all aspects of the biology of Malagasy bryophytes.

The project began in September 2017 with the important task of specimen sorting and digitization of collection information at the partner herbaria (MO, PC, TAN). Recently an identification workshop was conducted at the Antananarivo herbarium (TAN), Madagascar in September 2018. This workshop resulted in seven new reports for the Malagasy bryoflora (Candollea, in press). Taxonomic information is also being added to the XPer3 knowledge database system, allowing interactive species identification as well as flexible storage of taxa and characters as new information is gathered on the flora. We present here our very preliminary results in order to promote dynamic exchanges on Malagasy bryology.

Keywords: Madagascar, bryophytes, herbarium collection, systematic, knowledge database

The Hornworts (*Anthocerotophyta*) and thalloids liverworts (*Marchantiophyta*) of Cuba: challenges and perspectives

Yoira Rivera Queralta

Eastern Center of Ecosystems and Biodiversity (BIOECO). Ministry of Science, Technology and Environment (CITMA). Santiago de Cuba, Cuba. email: yoira@bioeco.cu

Abstracts

The main challenge of the botanists in Cuba is the upgrade of the bryologist studies. Presently work analyses the state of the knowledge of the hornworts and thalloids liverworts (*Marchantiophyta*) in the island, based on the review of National Herbarium (BSC), specialized literature, Databases of National (BSC) and International Herbarium (NY, MGB). The hornworts and talloids liverworts registered for Cuba are listed. The ecology, geographical distribution and state of conservation of the species are analyzed. About 60 species of hornworts and liverworts are recorded, 5% of the totals are endemic and 10% are threatened. The species registered for the island mostly present a dubious taxonomic status in response to the new World bryophyte Checklist. The hornworts and liverworts thalloids live in Cuban fragile ecosystems. This study sets the basis for the publication of the Checklist of Bryophytes of Cuba, as well as aims to awaken the interest to collaborate in future briological investigations on the island.

Key words: Cuba, hornworts, liverworts, *Marchantiophyta, Anthocerotophyta*. Thematic session: Biodiversity & Biogeograhy

Information for submission of abstracts to the Bryology 2019 meeting. Deadline: 31 May 2019

Presentation type: ??Oral Presentation, Poster Presentation

Assessing the resistance to lead and accumulation capacity of shoots, spores and adult plants to lead in four common moss species

<u>Alejandro R. Gijón¹</u>, Nagore G. Medina² & Belén Estébanez²

¹ Universidad Rey Juan Carlos, Móstoles (Madrid), C/ Tulipán s/n 28933 (<u>alejandro.rgijon@gmail.com)</u>; ² Universidad Autónoma de Madrid, Madrid (Madrid), C/Darwin 2 28049 (ngmedina@gmail.com, belen.estebanez@uam.es).

Mosses are frequently used as bioindicators because they absorb nutrients (and pollutants) directly from the ambient moisture across their entire surface. However, the effects of pollutants on their growth and development is not fully understood. For instance, we know little about the effective doses in different species, patterns of response or the ability of diverse moss species to exclude pollutants. In this study, we assess the sensitivity to lead and its accumulation on four common species of Iberian mosses with contrasted sensitivity to urban pollution: *Homalothecium aureum*, *Hypnum cupressiforme*, *Ptychostomum capillare* and *Syntrichia ruralis*.

We tested four lead nitrate doses $(0-10^{-3} \text{ M})$ on *in vitro* cultures of spores, shoot tips and adult stages of all those four species. Both spores and tips were cultivated directly in liquid medium with the corresponding lead dose. In adult stages, mosses were irrigated twice a week for 2 months with the different lead doses. In germination studies, we recorded survival and developmental stage of the spores, for shoot tips we recorded survival and damage rates. Finally, for adult populations we assessed damage using semiquantitative and quantitative measures and analysed its accumulation at the highest lead dose (10^{-3} M) using SEM/EdX.

The four species were more resistant to lead than anticipated. Lethal effects were evident only at the highest dose (10^{-3} M). Furthermore, a low but significant number of shoot tips and protonemata of *H. aureum* survived at the highest dose, and *S. ruralis* was able to produce sporophytes under all the doses. The SEM/EdX analysis showed that the mosses here analysed presented frequent lead aggregates on their surface, but were able to block effectively the entrance of this metal to the tissues. These results point at an exclusion mechanism, which allows them to survive at unexpectedly high lead doses, and that could seriously affect biomonitoring inferences of environmental lead pollution from metal levels found in living moss tissues.

Keywords: heavy metals, lead, pollution, biomonitoring, cultures.

Micro-morphology and anatomy of the peristome in the Dicranales: exploring the use of Technovit 7100 Methacrylate embedding resin for microtome sectioning

Mathilde Ruche^{1,2}, Michelle J. Price^{1,2}, Fred W. Stauffer^{1,2}

¹ Department of Botany and Plant Sciences, University of Geneva, Quai Ernest-Ansermet 30, 1211 Geneva, Switzerland (Mathilde.Ruche@unige.ch); ² Lab. of Plant Systematics and Biodiversity, Conservatory and Botanical Garden of the City of Geneva, Ch. de l'Impératrice 1, 1292 Chambésy, Geneva, Switzerland.

Peristome characteristics in mosses have long been used for classification at the higher taxonomic ranks. The complex architecture of the peristome provides traits that are also of importance at lower taxonomic levels. The micro-morphology and anatomy of peristomes within and across the Dicranales will be explored, with the aim of elucidating evolutionary patterns. Peristome architectural features may provide a new set of characters that can provide insights into the deeprooted problems of clade resolution and taxonomy in this monophyletic group of plants. The first objective was to elaborate protocols and test techniques that would allow for detailed observations and the comparison of microscopic features within sections of the peristome of Dicranum scoparium Hedw. and selected taxa from Dicranum Hedw. and the Dicranales. A protocol for sample preparation, widely used in plant tissue observations, was adapted to the study of the peristome. Technovit 7100 is a commercial product based on glycol methacrylate (GMA) resin that is used for embedding specimens before microtome sectioning. For the peristomes, very thin (3 to 10 μ m) longitudinal sections of the peristome and peristome insertion region were made revealing the cellular patterns and the intricate architecture of the peristome teeth and peristome-capsule wall region. This embedding and sectioning technique has many advantages in histology as the sample can be easily orientated in the transparent resin before sectioning where it can be serially sliced into thin sections allowing for high resolution imaging. The protocol and images obtained are presented along with a discussion on the preliminary observations and findings.

Keywords: Sporophyte, anatomy, micro-morphology, peristome, GMA embedding, mosses.

Presentation type: Poster Presentation, Oral Presentation

Antioxidant enzymatic analysis on bryophyte *Syntrichia ruralis* [Hedw.] in semi-arid sandy grassland, Hungary

<u>Ruchika</u>^{*1}, Evelin Ramóna Péli² Zsolt Csintalan¹

¹Institute of Botany and Ecophysiology, Faculty of Agricultural and Environmental Sciences, Szent István University, 2100 Gödöllő, Pater karoly utca. 1.Hungary

²Department of Botany, University of Veterinary Medicine, 1078 Budapest, István utca. 2. Hungary *Corresponding author: ruchika.110291@gmail.com

Bryophytes are poikilohydric and act as an experimental model plants to study the mechanism of desiccation tolerance. However, there has been little is known about biochemical activities in non-vascular cryptogram. The objective of this study was to examine the antioxidant enzymatic activities (CAT, APX, POD) during rehydrated and air-dried states in *S.ruralis* to understand how this plant survives in different seasons.

Moss cushions were collected from semi-arid grasslands near Bócsa-Bugac, Kiskúnság region in Hungary from the two microhabitats based on orientation of the sandy dunes; north-east (NE) and southwest (SW) directions respectively in spring, summer, autumn and winter 2018.

Our results showed the statistical differences in the mean values of CAT, APX and POD antioxidant enzymatic activities ($p \le 0.05$) between the microhabitats in rehydrated state except CAT and APX values in air-dried state. However, the mean values of CAT, APX and POD were found to be statistically significant within each pair of seasons for both states. In rehydrated state, higher CAT, APX and POD values in summer, autumn and spring respectively whereas in air-dried state only in autumn season.

In conclusion, NE slope showed higher antioxidant enzymatic activities as compare to SW slope in both rehydrated and air-dried states and has different responses in each season. Differences in the antioxidant activities of slopes can refer different adaptation level on desiccation tolerance under even close microhabitats. Depending on the exposure, various recovery level of *S.ruralis* also can be hypothesized after rehydration.

Keywords: *Syntrichia ruralis*, Desiccation tolerance, Catalase, Ascorbate peroxidase, Guaiacol peroxidase.

Physcomitrella patens dehydrin, PpDHNA, acts like "chaperone" by conffering protection against stress effects through protein stability enhancement

<u>Ruibal C¹</u>, Castro A and Vidal S¹

¹Laboratorio de Biología Molecular Vegetal, Facultad de Ciencias, Universidad de la República. Iguá 4225, CP 11400. Montevideo, Uruguay. <u>cruibal@fcien.edu.uy</u>

Drought tolerance was an ancient adaptation that must be considered to have played an essential role in colonization of the terrestrial environment. The most primitive of extant land plants, the bryophytes may provide resources as to how this adaptation was achieved. *Physcomitrella patens*, the model bryophyte, is highly tolerant to dehydration, salinity and other abiotic stress factors, and thus also a model for stress tolerance. Our objective is to better understand drought and salt stress tolerance in *Physcomitrella* so as to generate tools for increasing abiotic stress tolerance in crops. We studied the functional importance of a *Physcomitrella* gene encoding a protein from the dehydrin family, PpDHNA. Knockout mutant of this gene exhibited reduced tolerance to osmotic, salt and sugar stress.

We studied the role of ABA in dehydrin protein stability, as we have previously shown that PpDHNA is rapidly degraded upon recovery of stress. Our results suggest a role for ABA in dehydrin protein stability. Furthermore, *ppdhnA* knockout mutant is unable to recover from exposure to protein denaturation inducing agent DTT, and in vitro activity of this protein prevents chemical or heat denaturalization of enzymes.

A comparative study of corticolous bryophytes on three-needle pine and oak tree in Chiang Dao wildlife sanctuary, Chiang Mai province, Northern Thailand.

<u>Nilobon Sevanakun¹</u>, Prakasit Rawiwan², Sattha Ghunthong² and Sahut Chantanaorrapint¹ ¹Department of Biology, Faculty of Science, Prince of Songkla University, Hat Yai, Songkhla, 90112, Thailand (nilobon.sev@gmail.com); ²Chiang Dao wildlife sanctuary, Chiang Dao district, Chiang Mai Province 50170, Thailand

Previous studies in pine forest have shown that the most important phorophyte attributes in determining corticolous bryophytes species is bark features. However, no study on this topic has been addressed in this country. Therefore, this study aimed to compare the richness, community and life form of corticolous bryophytes that occur on two majority trees with different bark characteristics at pine-oak forest, Chiang Dao wildlife sanctuary, Chiang Mai province, Thailand. The trees were three-needle pine (Pinus kesiya Royle ex Gordon) and oak (Fagaceae). Twenty-four trunks of each phorophyte were selected. Bryophytes were collected from tree trunk at 1-1.5 m height. In addition, bark characteristics of each phorophyte were examined. The results of bark characteristics show that bark pH and bark water-holding capacity of oak were significantly higher than threeneedle pine. Nineteen species were recorded on three-needle pine and 35 species were found on oak. Ochrobryum subulatum Hampe was found only on three-needle pine, whereas 17 species were found only on oak. Species richness ranged from three to eleven species and four to fourteen species on three-needle pine and oak, respectively. Bryophytes community on three-needle pine was dominated by Leucobryum aduncum var. scalare (Müll. Hal. ex M. Fleisch.) A. Eddy, but a distinctive species on oak was Wijkia surcularis (Mitt.) H.A. Crum. The study of life form showed that cushion was the highest cover on three-needle pine whereas the most widespread life form on oak was weft. Our results found that species richness and cover of life forms appeared to relate to the moisture conditions and acidity of the tree bark. These results helped to explain some differences of corticolous bryophyte species among host trees in pine-oak forest, Thailand.

Keywords: bryophyte community, bryophyte ecology, epiphyte, pine forest

The research progress and prospect of bryophytes in Tibet

SHAO Xiaoming¹ * FAN Yingjie¹ LIU Ling¹ JIANG Xinyuan¹ Wang Li¹ SONG Xiaotong¹ KOU jing² SONG Shanshan¹ JIANG Yanbin³

1 Beijing Key Laboratory of Biodiversity and Organic Farming, College of Resources and

Environmental Sciences, China Agricultural University, 100193 Beijing, China

2 School of Life Sciences, Northeast Normal University

3 College of Resources and Environment, Huazhong Agricultural University corresponding author:shaoxm@cau.edu.cn

Abstract: Tibet (E 78°25′-99°06′, N 26°50′-36°53′) lies on the southwest of the Qinghai-Tibet Plateau. The region covers more than 1.2 million square kilometers and the average altitude is more than 4000 m, has the obvious vertical climate characteristics. In addition, it has low oxygen content, strong solar radiation and low temperature. Rainfall decreases from southeast to north. Its complex landforms, physiognomy, and climate create a special heterogeneous environment for bryophytes.

In the early stage, William Mitten (1859) and Chen Pan-Chieh (1962) formulated the bryophytes in Tibet. From 1952 to 1979, more than 7000 specimens were collected in many scientific investigations in Tibet. After identification and classification, BRYOFLORA OF XIZANG was published in early 1985, laying a foundation for the study of bryophytes in Tibet. In the past decade, Bryologists have done a lot of work in the area and a total of 1,027 species of bryophyte have been recorded, belonging to 307 genera of 90 families

28 genera and 132 species of Pottiaceae were indentified here. We have studied the spatial distribution of it. *Didymodon* and *Bryoerythrophyllum* are its dominant genuses, the main environmental variables affecting their spatial distribution were temperature and altitude. In the context of climate warming, the two distribution centers will be mainly in semi-arid areas in the future and are about to move to higher altitudes and higher latitudes.

Studies on the morphological traits of bryophytes found that the morphological traits of *Bryum argenteum*, *Didymodon constrictus* and *Didymodon rigidulus* are different in adapting to the high-cold-drought environment in Tibet, revealing that different species have different response strategies to environmental change.

Future research should pay more attention to the physiological traits and biochemical stoichiometry of bryophytes, contact molecules and microorganism, to explore the adaptation strategies of bryophytes in different environments, and provides scientific basis for ecosystem service function assessment in Tibet and global climate change.

Key words: Tibet; bryophytes; species diversity; spatial distribution; functional traits

This work was financially supported by Natural Science Foundation of China (Grant No. 41771054, 31570474)

International Association of Bryologists (IAB), International Molecular Moss Science Society (iMOSS) and Sociedad Española de Briología (SEB) 2019 Conference, 9-12 July 2019, Royal Botanic Garden (CSIC), Madrid, Spain

Presentation type: Oral Presentation, Poster Presentation

Bryophytes of submontane calcareous beech forests on dolomites of Central Slovakia

Pavel Širka¹, Mariana Ujházyová¹, Karol Ujházy¹

¹ Department of Phytology, Faculty of Forestry, Technical University in Zvolen, T.G. Masaryka 24, 960 01 Zvolen, Slovakia (pavel.sirka@tuzvo.sk)

In the last century, an intensive phytosociological research on beech forest vegetation has been carried out in Slovakia. However, many regions are still unexplored and, moreover, bryophytes are often omitted in phytosociological relevés. In this paper, we analyzed bryophyte species richness and composition in relation to elevation, xericity, slope, cover of bare rocks, cover of herb, shrub and tree layer, height of tree layer and vegetation unit (association), as well as traits of the recorded bryophytes regarding their ecological and habitat preferences in forest ecosystems dominated by European beech (Fagus sylvatica) in the hilly region around Banská Bystrica built by dolomite rocks and covered by rendzina soils. The analysis was based on 67 bryophyte species (59 mosses and 8 liverworts) recorded in 45 phytosociological relevés from 400 m² plots. For the statistical analysis between bryophyte species richness and composition and selected environmental variables Spearman's rank correlation coefficient, ANOVA and CCA were used. Ellenberg's indicator values and BRYOATT database were used to compile trait information. The most frequent species in the relevés was Tortella tortuosa, followed by Hypnum cupressiforme, Brachytheciastrum velutinum and Ctenidium molluscum. Average species richness of bryophytes was 8, compared to 39 species of vascular plants in the forest understory. Bryophyte species richness was negatively affected by xericity and positively by cover of rocks on soil surface. Species composition was affected significantly by xericity, cover of bare rocks and cover of tree layer. The majority of the taxa (88.1%) are adapted to moderately moist soils, and roughly two thirds prefer moderately to strongly basic soils but 14% are acidophilous. Semi-shade species are the most numerous (71.2%) but a significant portion (23.7%) are semi-light or found in full light, whereas 50.9% are indicators of colder climate. Proportionally, species show habitat preference mostly to hard rocks (84.1%), living wood (66.7%) or soil (52.4%). Our results indicate that in the studied area, bryophyte species composition is mostly influenced by moisture availability (conditioned by xericity and cover of tree layer) and the amount of bare rocks greatly contributes to the overall diversity of available substrates and thus to a higher species richness of bryophytes.

Keywords: bryophyte traits, species richness and composition, environmental variables, *Fagus sylvatica*, Banská Bystrica

Effect of peatland degradation on bryophyte species composition in Latvia and Lithuania

<u>Liga Strazdina¹</u>, Jurate Sendzikaite^{2,3}, Mara Pakalne¹, Agnese Priede¹, Leonas Jarasius², Zydrunas Sinkevicius², Nerijus Zableckis²

 ¹ University of Latvia, Botanical Garden, Kandavas Street 2, Riga, LV-1083, Latvia (liga.strazdina@lu.lv);
² Lithuanian Fund for Nature, Algirdo Street 22-3, Vilnius, LT-03218, Lithuania;
³ Nature Research Centre, Zaliuju Ezeru Street 49, Vilnius, LT-08406, Lithuania (jurate.sendzikaite@gamtc.lt)

Bryophytes are pioneer species that occupy degraded sites, both after natural disturbances (e.g. fire) and human impact (e.g. logging). Post-harvested peatlands can be inhabited by bryophyte species that are adapted to extreme conditions such as drought, exposure to high sun radiation, wind erosion, etc. Still, bryophyte richness is higher in intact or less affected peatlands – raised bogs, transition mires and fens with different ecological niches. The role of bryophytes, especially Sphagnum, in mires involves also peat formation and carbon sequestration. Climate change mitigation by reducing greenhouse gas (GHG) emissions, low groundwater fluctuations are only some functions that are strongly related to processes in intact peatlands. To study bryophyte species composition in peatlands with different management history, data were collected in eight sites: three peatlands with minor drainage impact in Latvia and five significantly drained, post-harvested peatlands in Lithuania. Vegetation cover (bryophytes, lichens, herbs, shrubs, trees) and physical parameters (degree of site degradation, soil moisture) were recorded in 269 plots in 2017–2018 during the LIFE Peat Restore project (LIFE15 CCM/DE/000138). Additionally, vegetation data together with environmental parameters were used for indirect GHG emission assessment applying the recently developed GEST (Greenhouse Gas Emission Sites Types) approach. In all sites, 382 species were recorded, including 85 bryophyte species (10 liverworts and 75 mosses, including 19 Sphagnum species). Bryophyte species richness showed moderate positive correlation with total species richness (r_s =0.53, p<0.001), weak correlation with moisture (r_s =0.34, p<0.001) and herb cover/species richness (r_s =0.24, p<0.001; r_s =0.28, p<0.001, respectively). Bryophyte species richness was best explained by total number of species per plot, bare peat and bryophyte species cover and was site-specific. Species composition significantly differed between degraded and near-natural sites. From all recorded bryophytes, 24 species were related to one of four previously defined degradation classes using indicator-species analysis. Campylopus introflexus, Pleurozium schreberi, Pohlia nutans, Polytrichum spp. were indicators of the most degraded sites, while Cephalozia bicuspidata, Dicranum bonjeanii, Mylia anomala, Sphagnum angustifolium were found only in sites with minor drainage impact. The results suggest that bryophytes can be used as indicators to assess the degradation of peatlands.

Keywords: degraded peatlands, bryophyte species composition, indicators, GEST approach

Codonoblepharon forsteri (Orthotrichaceae) has its most important population in Southwestern Spain

Terroba, A.G.¹, Draper, I.^{1,2}, Mazimpaka, V.^{1,2} Garilleti, R.³, & Lara, F.^{1,2}

¹ Departamento de Biología, Facultad de Ciencias, Universidad Autónoma de Madrid, c/ Darwin, 2. E-28049, Madrid; ² Centro de Investigación en Biodiversidad y Cambio Global (CIBC-UAM), c/ Darwin, 2. E-28049, Madrid. <u>isabel.draper@uam.es</u>; <u>vicente.mazimpaka@uam.es</u>; <u>francisco.lara@uam.es</u>; ³ Departamento de Botánica y Geología, Facultad de Farmacia, Universidad de Valencia, Avda. Vicente Andrés Estellés s/n. E-46100, Burjassot. <u>ricardo.garilleti@uv.es</u>

Codonoblepharon forsteri (Dicks.) Goffinet is a peculiar moss from the Atlantic area of Europe, North Africa and Macaronesia. Despite its extensive range, it is a rare species in practically all the countries in which it has been found. This seems to be due to the fact that it is a very specialized and highly stenoicous epiphytic moss, with very specific ecological requirements: it invariably lives on large trees, limited to runoff areas of the bark associated with holes, scars, tree forks and other well-lit, humid and nutrient-rich situations (tree stumps, exposed roots, etc.). The rarity of this moss could also be due to demographic reasons, since the species shows very low densities throughout its range.

In the Iberian Peninsula, *Codonoblepharon forsteri* has been reported from scattered localities, mostly in the northern and western areas. Without exception, only one or very few colonized trees are known in each locality. Thus, only 10 colonized trees have been found in all the 9 localities reported for Portugal (Garcia, 2006) and almost all the 16 known Spanish localities showed each only one colonized tree (Heras Pérez *et al.*, 1997).

In the present communication, a large population of *Codonoblepharon forsteri* is reported from Ronda (Andalusia, Southern Spain). We provide data on the size of the population, the distribution of the colonies in the surveyed area, and the environmental characteristics, at different ecological levels (forests, phorophyte and microtope), in which the moss grows. Local practices of forest management that affect and can threaten this extraordinary population are also identified.

Keywords: Iberian Peninsula, Population size, Threatened moss, Zygodon forsteri,

References:

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Moss-water-interactions: Effects of dehydration and hydration on moss surface structures

Sonja M. Thielen¹, Martin Ebner¹, Tatiana Miranda^{1,2}, James H. Nebelsick¹, Anita Roth-Nebelsick³

¹ Department of Geosciences, Eberhard-Karls-Universität Tübingen, 72074 Tübingen, Germany (sonja.thielen@uni-tuebingen.de); ² Senckenberg Centre for Human Evolution and Paleoenvironment (S-HEP-Tübingen), 72074 Tübingen, Germany; ³ State Museum of Natural History Stuttgart, 70191 Stuttgart, Germany

Mosses as poikilohydric plants do not actively regulate their water content, which is in equilibrium with environmental humidity. They are able to survive long drought periods by several adaptations, leading to a pronounced desiccation tolerance. Photosynthetic activity, however, requires hydrated tissue. It is long known that available water is taken up via the whole moss surface. On the other hand, mosses need to avoid the formation of a water film covering the assimilating tissue and hampering photosynthesis by acting as a diffusion barrier. Despite the vital importance of efficient water absorption, information on moss surface interactions with water are sparse. In this study, four epiphytic moss species were analysed with respect to surface structure response to the dehydrated and hydrated state. Samples were collected at four sites in the district of Tübingen, Germany, during October and November 2018. Samples were kept dry for at least 30 days. To study hydrated phylloids, samples were submersed in distilled water for 10 min. To conserve cell structures in a dehydrated or hydrated state, dry and hydrated samples were shock-frozen with liquid nitrogen, directly followed by lyophilisation for 24 h. Subsequently, the morphological response towards the different water conditions was analysed using a scanning electron microscope. Dry and turgescent state differed in cell width, cell length and surface structure. Also, species reaction on hydration and dehydration was distinct from one another in the way surface structures changed. In a next step, the analysed structures will be further examined with respect to adaptive significance and a possible application potential.

Keywords: Water relations, surface structures, epiphytes

Trace elements concentrations in Greece using Hypnum cupressiforme Hedw. as biomonitor

<u>Evdoxia Tsakiri¹</u>, Chrysoula Betsou², Nerantzis Kazakis³, Konstantinos Eleftheriadis⁴, Evangelia Diapouli⁴, Marina Frontasyeva⁵, Alexandra Ioannidou²

¹Laboratory of Systematic Botany & Phytogeography, Department of Botany, School of Biology, Aristotle University, Thessaloniki 54124, Greece (tsakiri@bio.auth.gr);² Nuclear and Elementary Particle Physics Laboratory, Physics Department, Aristotle University, Thessaloniki 54124, Greece (chbetsou@physics.auth.gr; anta@physics.auth.gr); ³ Division of Hydrogeology, Geology Department, Aristotle University, Thessaloniki 54124, Greece (kazakis@geo.auth.gr); ⁴ Institute of Nuclear & Radiological Sciences and Technology, Energy & Safety, NCSR "Demokritos", Athens 15341, Greece (elefther@ipta.demokritos.gr; Idiapouli@ipta.demokritos.gr);⁵ Frank Laboratory of Neutron Physics, Joint Institute for Nuclear Research, Dubna, Moscow Region, 141980, Russia (marina@nf.jinr.ru)

Data of the Greek participation to the European Surveys of Heavy Metals accumulation in Mosses are presented in this study (United Nations Economic Commission for Europe International Co-operative Programme on Effects of Air Pollution on Natural Vegetation and Crops). The area selected for study was Northern Greece (Districts of West, Central and East Macedonia and Thrace). The study aimed at mapping of heavy metals and toxic elements accumulation (Al, As, Cr, Fe, Ni, Sb, V, Zn) in the moss Hypnum cupressiforme Hedw., reflecting wet and dry atmospheric deposition. Moss samples were obtained from 95 sites situated in Northern Greece, covering ca 42,000 km². Preliminary moss samples preparation, chemical analysis, quality control, etc., followed the ICP Vegetation Monitoring Manual requirements and the procedure was done in Frank Laboratory of Neutron Physics, Joint Institute for Nuclear Research (Dubna, Russian Federation). Hypnum cupressiforme samples were analyzed using the Neutron Activation Analysis (NAA) technique and the elements concentrations were determined. The chemical composition database of the moss samples was further used for the application of source apportionment by Positive Matrix Factorization (PMF). In total thirty chemical elements were used for source apportionment (Na, Mg, Al, Si, Cl, K, Ca, Sc, Ti, V, Cr, Mn, Fe, Co, Ni, As, Br, Rb, Sr, Mo, Cd, Sb, Cs, Ba, La, Ce, Tb, Hf, Ta and Th). The big sampling density provided information about the elemental deposition from the atmosphere to terrestrial systems over the region of Northern Greece. The source apportionment results revealed contribution from five sources: (1) Soil Dust, (2) Aged Sea Salt, (3) Vehicular Traffic, (4) Heavy Oil Combustion and (5) Mining Activities. Soil Dust displayed the highest contribution to the measured metal concentrations among all other sources. Additionally, two areas in Northern Greece, the Megali Panagia area and the Ptolemaida-Amyntaio area were well connected with specific sources; for Megali Panagia there was high contribution of road dust and Mn-rich sources which are related to the nearby existing mining activities, while the Ptolemaida-Amyntaio area was correlated with the heavy oil combustion source related to the lignite power plants located in the wider area.

Keywords: Atmospheric deposition, Hypnum cupressiforme, heavy metals

The role of prenyltransferases in the thallus development of Marchantia polymorpha

<u>Lia R. Valeeva</u>¹, Yoshihiro Yoshitake², Noriyuki Suetsugu², Ryuichi Nishihama², Takayuki Kohchi², Margarita R. Sharipova¹.

¹Institute of fundamental medicine and biology, Kazan (Volga region) federal university,Kazan 420021, Russia (<u>lia2107@yandex.ru</u>), ² Graduate School of Biostudies, Kyoto University, Kyoto 606-8502, Japan.

Protein prenylation is one of the most important post-translational modifications that plays an important role in processing of normal cellular mechanisms, such as signal transduction, polar cell growth, membrane and cell wall modifications. All of these processes are an integral part of the functioning of multicellular organisms. Knockouts in prenyltransferase genes can lead to dramatic changes in plant growth and development, up to the loss of the ability to form normal multicellular tissues. Multicellularity transition is one of the most important events in the evolution of living organisms, but most facts about this event remain to be unknown. Using of plants, especially evolutionally ancient groups like Bryophytes, allows us to find out new regulatory mechanisms involved in development of multicellular organisms.

We investigated the role of farnesyltransferase and geranylgeranyltransferase genes in the *Marchantia polymorpha* multicellular thallus' development. We obtained knockout lines for three genes of prenyltransferases subunits: a gene of the alpha-subunit PLP (Mapoly0093s0047), common for farnesyl- and geranylgeranyltransferase; a gene of farnesyltransferase beta-subunit ERA (Mapoly0123s0027) and agene of geranylgeranyltransferase I beta-subunit GGB (Mapoly0010s0084). Knockout plants for each genes were generated by CRISPR/Cas9 technology. It was shown that *Marchantia* lines with mutations in the sequence of the alpha subunit gene ($\Delta p l p$) formed callus-like dense tissues that were composed of round-shaped cells of various sizes and easily disintegrated into separated fragments. Although $\Delta p l p$ mutants retained the ability to aggregate into tissue, they were not capable for forming a normal thallus that undoubtedly indicates the important role of prenyltransferases in the development of tissues, cell differentiation and the formation of multicellular thallus.

The work was supported by the Russian Science foundation grant 18-74-00112.

Keywords: prenyltransferases, Marchantia polymorpha, development, multicellularity

Campylopus introflexus colonization in the Atlantic secondary dunes of the Iberian Peninsula.

<u>Z. Varela</u>^{1,2,*}, S. Lobo Dias², C. Branquinho²

¹ Ecology Unit, Dept. Functional Biology, Facultade de Bioloxía, Universidade de Santiago de Compostela, 15782 Santiago de Compostela, Spain (<u>zulema.varela@usc.es);</u>² Centre for Ecology, Evolution and Environmental Changes (cE3c), Faculdade de Ciências, Universidade de Lisboa, 1749-016 Lisbon, Portugal.

Campylopus introflexus (Hedw.) Brid. is considered one of the 100 worst alien invasive species in Europe. Thanks to its high ecological tolerance, this moss species has spread throughout all European countries especially in places subject to a high anthropic influence. Despite its high tolerance C. introflexus prefers dry, sandy, nutrient-poor sites with little herbaceous vegetation which make the coastal dunes an optimal habitat for the emergence and spread of this invasive species. The presence of bryophytes in dunes is common thanks to their poikilohidric features where they can play an important role in stabilization of dune surfaces. However, the existence of a threatening species such as C. introflexus can endanger the fragile equilibrium of the plant communities in these coastal habitats. This work intends to assess the causes of the presence of Campylopus introflexus on secondary dunes in relation with several biotic and abiotic factors including bryophyte and vascular plant communities. For this, we sampled transects parallels to the coastline and identified bryophytes and other vegetation type using the point-intercept method in 15 beaches along a climatic gradient in the western coast of the Iberian Peninsula. The results showed that C. introflexus is only present when aridity decreases, and in fact its presence is strongly correlated with precipitation variables. In addition, at intermediate values of aridity, it seems that there is a competition between Campylopus i. and the common native moss Pleurochaete squarrosa (Brid.) Lindb. Negative correlations were also found between Campylopus i. and perennial vegetation cover and diversity, that can be due to anthropic disturbance. These findings suggest that the level of aridity, vegetation cover and diversity and anthropic influence can contribute to the colonization of secondary dunes by the alien invasive moss Campylopus i.

Keywords: Aridity, Bryophytes, Coastal dunes, Invasive species

Acknowledgments: This work was supported by the project UNITED -Unificando nichos, interacciones y distribuciones: Un entorno teórico común para dinámica de rangos geográficos y coexistencia local" funded by Spanish Goverment and by the project ChangeTracker, FCT-PTDC/AAG-GLO/0045/2014. ZV was supported by a PostDoc research grant awarded by the Autonomous Government of Galicia (Spain) and SL was supported by PhD Grant FCT-PD/BD/114438/2016.

Impact of transposable elements in the generation of phenotypic variability in *Physcomitrella* patens

Pol Vendrell Mir¹, Florence Charlot², Fabien Nogué², Josep Casacuberta².

¹ Center for research in Agricultural Genomics (CRAG), 08193 Cerdanyola del Vallès, Barcelona, Spain (<u>pol.vendrell@cragenomica.es</u>); ²₄Institut Jean-Pierre Bourgin(IJPB)-INRA,78026 Versailles, France

Transposable elements (TEs) comprise 57% of the genome of Physcomitrella patens (Lang et al. 2018). Using a recently published RNA seq gene atlas of the P. patens genome (Perroud et al. 2018) we have been able to assemble the complete transcripts of 6 active families of transposable elements, including retrotransposons (LTRs-retrotransposons and LINEs) and DNA transposons. These transposable elements show different pattern of expression in tissues or stress conditions. These results suggest that different TE families are active in Physcomitrella patens. We have searched for TE polymorphism in four ecotypes of Physcomitrella patens using resequencing data of these ecotypes. The results suggest that these families have been recently active and may be at the origin of some of the phenotypic differences observed in these ecotypes. As a first approach we are analysing the patterns of expression of genes with a TE polymorphisms in the proximal upstream region of the gene. As an example, we found that the expression of a transcription factor highly expressed during rhizoid development is downregulated in Villersexel as compared to Gransden and that this decrease of expression is potentially due to an LTR-retrotransposon insertion. We have generated a knock out of the corresponding gene by CRISPR/Cas9 in Gransden and show that rhizoids development in this mutant is reminiscent of the one observed in Villersexel, suggesting that the LTR-retrotransposon insertion, and the decrease of expression of this gene, could be at the origin of the differences observed in rhizoid development between Gransden and Villersexel. In order to confirm this hypothesis we are performing locus replacement between the two varieties in order to check the hypothesis that insertion of the TE is indeed involved in the phenotype observed.

Keywords: Transposable elements, variability, transcription

Lang *et al.* (2018) *The Plant Journal* 93.3: 515-533. Perroud *et al.* (2018) *The Plant Journal* 95.1 : 168-182. **Con formato:** Sin subrayado, Color de fuente: Automático

Con formato: Sin subrayado, Color de fuente: Automático

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New insights in taxonomy and phylogeny of liverworts

<u>Rui-Liang Zhu¹</u>, Lei Shu¹, You-Liang Xiang¹, Chao Shen¹, Xin-Jie Jin¹

¹ Bryology Laboratory, School of Life Sciences, East China Normal University, 500 Dongchuan Road, Shanghai 200241, China (rlzhu@bio.ecnu.edu.cn)

The liverworts, one of three early diverging extant land plant lineages, are represented by over 7000 species in 371 genera. Although much progress in molecular phylogenetics has been made in recent years, the division of liverworts into natural genera has not yet been satisfactorily solved. The phylogenetic relationship of many enigmatic genera is still not clear. The main objective of this study is to resolve the phylogenetic relationship of key groups of liverworts using molecular data including complete chloroplast genomes and transcriptomes. Our new data support the current higher-order classification of liverworts. The sporophyte of *Mizutania* Furuki et Z.lwats., an extraordinary genus previously placed in the simple thalloid liverworts, is found for the first time. On the basis of morphological and molecular data, several enigmatic genera such as *Aphanotropis* Herzog, *Calatholejeunea* K.I.Goebel, *Cephalolejeunea* Mizut., *Dactylophorella* R.M.Schust., *Sinomylia* P.C.Wu et al., are reduced to be new synonyms. Two new genera from Oceania and tropical Asia are proposed. The status of several genera of Lejeuneaceae with rampant homoplasy, rapid radiations, and presence of neoteny, e.g. *Tuyamaella* S.Hatt. and *Schusterolejeunea* Grolle, is greatly challenged. A more intensive analysis of chloroplast genomes and transcriptomes from extended samples on a broader scale may provide better phylogenetic resolution.

Keywords: classification, genome phylogeny, Jungermanniopsida, new genus, new synonym.

Results of long-term bryophyte monitoring of dry rocky grasslands in Hungary

<u>E. Szurdoki¹</u>, P. Ódor², B. Papp¹

¹Botanical Department, Hungarian Natural History Museum, H-1088, Budapest, Baross u. 13, Hungary (szurdoki.erzsebet@nhmus.hu, papp.beata@nhmus.hu), ²Institute of Ecology and Botany, Hungarian Academy of Sciences, H-2163 Vácrátót, Alkotmány u. 2-4., Hungary (odor.peter@okologia.mta.hu)

The bryophyte vegetation monitoring is an ongoing project started in 2000 in the frame of the Hungarian Biodiversity Monitoring System and includes 50 sampling plots of various habitat types (wetlands, dry grasslands, saline-alkali areas and forests). The size of the permanent plots are 10 m x 10 m and twenty-five 0.25 m² sized quadrats are placed regularly at the corners of a grid of 2 m spacing within the plots. In each quadrat, the presence of bryophyte species is recorded. The recent presentation focuses on the changes in the bryophyte assemblages of dry rocky grasslands. The survey has been carried out in eight permanent plots by biennial sampling, and there are already 8-10 repetitions per plots. The species richness of dry rocky grasslands is high compared with the bryophyte assemblages of other habitat types surveyed. The species richness was between 11 and 28 in the plots. The species pool of the plots has been continuously increased; the mean species number per plot was 18 in the first year (between 11 and 30) and the mean cumulative species number was 32.5 in the last year (between 21 and 42). The mean number of new species in plots was 3.6 (between 0 and 8). The one-third part of all detected species per plots were present in each surveyed year, almost 60% of species occured at least half of releves and 17% of them were detected only once per plot. General structure of the assemblages of different years was analysed by PCoA using plot level frequency data. There were many fluctuations at bryophyte species level, but there were no trends at assembly level. To study the annual dynamics of the species, the relationships between mean annual increment and mean annual turnover were evaluated. Most of the species showed the dynamic expected on the base of their life strategy, but colonist species formed a very heterogeneous group. Surveyed dry grasslands seem to be stable. The differences in species richness and species composition in various years derives from their natural population dynamics.

Keywords: bryophyte assembly, dynamics, permanent plots

Information for submission of abstracts to the Bryology 2019 meeting. Deadline: 31 May 2019

Presentation type: Oral Presentation, Poster Presentation (underline the preferred type)

Abstract Title: Human impact on moss biodiversity in Omayed Biosphere Reserve (OBR), Egypt

El-Sakaty S.I.A.¹, Magdy M.², Abu-Elhamd M.F.¹

Corresponding author & presenter: Sahar_elsakaty@sci.asu.edu.eg

¹ Botany Department, Faculty of Science, Ain Shams University, Cairo 11566, El-Khalifa El-Mamoun Street, Abbassia, Cairo, Egypt. ² Genetic Department, Faculty of Agriculture, Ain Shams University, Cairo 11241, Hadayek Shubra, No. 68, Cairo, Egypt.

Omayed Biosphere Reserve (OBR) is located in the western north coast (Mediterranean – Ecosystem type) of Egypt. It is considered as a hub for moss spores carried from southern Europe to Egypt, where diaspores fall and grow but naturally fail to spread further in the surrounding Egyptian western desert, probably except in the Oasis because of the availability of relatively suitable microhabitats. where only six moss taxa out of the 34 recorded in OBR were reported from the Oasis. OBR contains 12% of Egypt's flora of flowering plants, endemic and rare animals and more than 15% of the bryoflora. After political disturbance of 2011, the human impacts increased; construction of touristic villages, intensive agriculture activity beside some Military actions resulted in major decrease in the numbers of all organisms. Repeated trips to OBR in winters of 2016, 2017 and 2018 showed the decrease in number of collected moss samples and taxa with noticeable occupation of their microhabitats by invasive flowering plants. Five taxa of Pottiaceae were repeatedly collected in all the trips and were identified morphologically and molecularly as *Tortella nitida, Tortula muralis, Trichostomum crispulum, Aloina brevirostris and Didymodon vinealis*. This work puts a flash spot that might attract attention of any concerned parties that OBR needs rapid intervention to save its unique biodiversity.

Keywords: Omayed Biosphere Reserve, Egypt, mosses, human impact