

USB - SECTION BIOLOGY

**INSTITUTE OF BIODIVERSITY AND
ECOSYSTEM RESEARCH – BAS**



11th SEMINAR OF ECOLOGY - 2018
WITH INTERNATIONAL PARTICIPATION

26-27 April 2018

**Програма/Program
Абстракти/Abstracts**



ORGANIZING COMMITTEE:

Chairmen:

Prof. STEPHKA CHANKOVA PhD-IBER-BAS
Assoc. Prof. ANNA GANEVA PhD-IBER-BAS

Chief Secretaries:

Prof. SPASIMIR TONKOV DSc -Sofia University
Assist. Prof. PETYA PARVANOVA PhD-IBER-BAS
Assist. Prof. KALINA DANOVA PhD-IOCCP-BAS

Committee members:

Assist. Prof. MARIA TODOROVA PhD-IBER-BAS
Assist. Prof. MARIA YOVKOVA PhD-IBER-BAS
Assist. Prof. MAYA KURTEVA PhD-IBER-BAS
Assist. Prof. RABIA SOUFI PhD-IBER-BAS
Assist. Prof. RADKA FIKOVA PhD-IBER-BAS
Assist. Prof. YANKA VIDINOVA PhD-IBER-BAS
Assist. TSVETA ANGELOVA-IBER-BAS
Biologist GALIA GEORGIEVA-IBER-BAS
Biologist GERGANA GEORGIEVA-IBER-BAS
Chemist ZHANA MITROVSKA-IBER-BAS
Engineer IVAN YANCHEV-IBER-BAS

SCIENTIFIC COMMITTEE:

Prof. BOYKO GEORGIEV DSc -IBER-BAS Prof.
ELISAVETA STOIMENOVA PhD-IPPG-BAS Prof.
MARIYANA LYUBENOVA PhD-Sofia University Prof.
RUMEN KALCHEV PhD -IBER-BAS
Prof. STEPHKA CHANKOVA PhD- IBER-BAS
Prof. SVETLANA BANCHEVA PhD-IBER-BAS
Prof. VLADA PENEVA PhD-IBER-BAS
Assoc. Prof. ASSEN ASSENOV PhD-Sofia University
Assoc. Prof. EMILIA VARADINOVA PhD-IBER-BAS
Assoc. Prof. GALINA RADEVA PhD-IMB-BAS
Assoc. Prof. MARGARITA TOPASHKA PhD-IBER-BAS
Assoc. Prof. MARINA STANILOVA PhD-IBER-BAS
Assoc. Prof. HRISTINA KALCHEVA PhD-IBER-BAS

The Seminar of Ecology is organized by Section Biology of the Union of Scientists in Bulgaria and the Institute of Biodiversity and Ecosystem Research, BAS, Sofia.

Many thanks for financial support of LKB-Bulgaria EOOD, Bulgap EOOD and Pensoft Publishers EOOD.



Contents:	
Program	1
Abstracts	11
Session I: Biodiversity and conservation biology	
▪ Lectures	11
▪ Poster presentations	21
Session II: Biotic and abiotic impact of the living nature and mechanisms of adaptation	
▪ Lectures	32
▪ Poster presentations	39
Session III: Ecosystem research, services and ecological agriculture	
▪ Lectures	46
▪ Poster presentations	48
Session IV: Landscape ecology	
▪ Lectures	51
▪ Poster presentations	53
Session V: Ecology and education	
▪ Lectures	53
▪ Poster presentations	54
Session VI: Other related topics	
▪ Poster presentations	56
Index	57



**11th ANNUAL
“SEMINAR OF ECOLOGY – 2018”
WITH INTERNATIONAL PARTICIPATION**

26-27 April 2018

PROGRAM

26 April 2018

8⁰⁰ - 9⁰⁰ Registration (IBER-BAS-2 Gagarin Str., Sofia)

9⁰⁰ – 9³⁰ Opening Ceremony

9³⁰ – 9⁴⁵ Family photo

9⁴⁵-10⁰⁰ "Seminar of Ecology - 11 years in the scientific skyline, first year on the Internet" a presentation on the first web site of the Seminar. *Galia Georgieva*

10⁰⁰-10³⁰ IBER-BAS *Assoc. Prof. Anna Ganeva*

THEMATIC SESSION

BIOTIC AND ABIOTIC IMPACT OF THE LIVING NATURE AND MECHANISMS OF ADAPTATION


Chairmen: Prof. Stephka Chankova and Prof. Snezhana Grozeva


Plenary presentations


10³⁰-10⁵⁰ INVASIVE ALIEN SPECIES – POTENTIAL CHEAP RESOURCES OF PLANT SUBSTANCES FOR MEDICINAL USE *Ekaterina Kozuharova, Iliana Krasteva, Aleksandar Shkondrov, Iliana Ionkova- PL02_01*


10⁵⁰-11¹⁰ EFFECT OF FUNGICIDES ON SOIL MICROBIAL COMMUNITY *Galina Radeva, Michaella Aleksova, Silvena Boteva, Anelia Kenarova- PL02_02*

Oral presentations

11¹⁰-11²⁰  CHARACTERISTICS OF PHOTOSYNTHETIC APPARATUS OF *ERYNGIUM MARITIMUM* L. AND *POLYGONUM MARITIMUM* L. - HALOPHYTIC PLANTS FROM DUNES OF POMORIE LAKE (BULGARIA) WITH DIFFERENT ADAPTIVE STRATEGY AGAINST SOIL SALINITY *Lilia Angelova, Albena Ivanova, Irina Kosakivska, Lilia Babenko, Svetlana Momchilova, Sabina Taneva, Albena Momchilova, Liliana Maslenkova- L02_01*

11²⁰-11³⁰  COMPARISON OF THE EFFECTS OF SALT STRESS ON TWO PAULOWNIA LINES *Martin Stefanov, Ekaterina Yotsova, Emilia Gesheva, Velmira Dimitrova, Yuliana Markovska, Snezhana Doncheva, Emilia Apostolova - L02_02*

11³⁰-11⁴⁰  MOLECULAR DETECTION OF *NOSEMA CERANAE* AND *NOSEMA APIS* IN BULGARIAN HONEY BEES *Iskra Yankova, Daniela Sirakova, Boyko Neov, Georgi Radoslavov, Peter Hristov- L02_03*

11⁴⁰-11⁵⁰  BIOLOGICAL AND STATISTICAL COMPARISON OF EXPERIMENTAL RESULTS- GOOD APPROACH FOR ANALYZING ADAPTIVE POTENTIAL OF GENETICALLY CLOSELY RELATED GENOTYPES *Tsveta Angelova, Valeria Simeonova, Zhana Mitrovska, Stephka Chankova - L02_04*

11⁵⁰-12⁰⁰ NURELLE D BIOACTIVITY DEPENDING ON THE TEST SYSTEM *Teodora Todorova, Petya Parvanova, Abdel-Tawab H. Mossa, Kostadin Kostadinov, Martin Dimitrov, Ivan Iliev, Zhana Mitrovska, Tsveta Angelova and Stephka Chankova - L02_05*

12⁰⁰-12¹⁰  IS THERE A RELATIONSHIP BETWEEN DNA REPAIR EFFICIENCY, HSP_s AND GENOTYPE RESISTANCE? *Daniela Miteva, Zhana Mitrovska, Nadejda P. Yurina, Stephka Chankova- L02_06*

12¹⁰-12²⁰ GENOTOXIC AND MUTAGENIC EFFECTS OF *NARCISSUS TRIANDRUS* L. TOTAL EXTRACT ON *CHLAMYDOMONAS REINHARDTII* *Petya Parvanova, Zhana Mitrovska, Boriana Sidjimova, Milena Nikolova, Strahil Berkov and Stephka Chankova- L02_07*

12²⁰-12⁴⁰ Discussion

12⁴⁰-13⁴⁰ Lunch

THEMATIC SESSION ECOLOGY AND EDUCATION

Chairmen: Prof. Spasimir Tonkov and Prof. Daniela Pilarska

Plenary presentations

13⁴⁰-14⁰⁰ SUCCESSFUL IMPLEMENTATION OF PROJECT-BASED LEARNING IN HIGHER EDUCATION: AN EXAMPLE FROM ECOLOGY *Kamelia Yotovska, Victoria Necheva- PL05_01*

Oral presentations

14⁰⁰-14¹⁰ THE SCHOOL EDUCATION – A CONDITION TO FORM POSITIVE ATTITUDE TOWARDS ENVIRONMENT OR ECOLOGY *Ekaterina Yovcheva, Veneta Kirkovska- L05_01*

14¹⁰-14³⁰ Discussion

THEMATIC SESSION LANDSCAPE ECOLOGY

Chairmen: Prof. Svetlana Bancheva and Prof. Mariyana Lyubenova

Plenary presentations

14³⁰-14⁵⁰-ACADEMICIAN ANASTAS ISHIRKOV'S CONTRIBUTION TO THE DEVELOPMENT OF BIOGEOGRAPHY IN BULGARIA (ON THE OCCASION OF 150 YEARS FROM HIS BIRTH) *Assen Assenov-PL04_01*

Oral presentations

14⁵⁰-15⁰⁰ - FIRST ATTEMPT OF A HIERARCHICAL CLASSIFICATION OF BENTHIC SEASCAPE UNITS IN THE BULGARIAN BLACK SEA *Iliyan Kotsev- L04_01*

15⁰⁰-15¹⁰- LAZARUS TAXA IN ANIMALIA KINGDOM *Borislav Grigorov, Assen Assenov-L04_02*

15¹⁰-15³⁰ Discussion

15³⁰-16⁰⁰-Coffee break

16⁰⁰-18⁰⁰ Poster sessions (*BIOTIC AND ABIOTIC IMPACT OF THE LIVING NATURE AND MECHANISMS OF ADAPTATION; ECOLOGY AND EDUCATION; LANDSCAPE ECOLOGY; OTHER RELATED TOPICS*) and Discussion

Chairmen: Assoc. Prof. Assen Assenov and Assoc. Prof. Galina Radeva

POSTER SESSION

BIOTIC AND ABIOTIC IMPACT OF THE LIVING NATURE AND MECHANISMS OF ADAPTATION


P02_01 EUROPEAN ROE DEER (*CAPREOLUS CAPREOLUS*) AS A BIOMONITOR FOR THE CURRENT ENVIRONMENT HEAVY METALS CONTAMINATION IN AN AGRICULTURAL REGION IN BULGARIA *Georgi Markov, Atidzhe Ahmed*


P02_02 INTERRELATIONSHIP BETWEEN FRESH AND DRY WEIGHT OF INTERNAL TARGET ORGANS OF ROE DEER (*CAPREOLUS CAPREOLUS*): APPLICATION FOR THE PURPOSES OF BIOMONITORING OF THE ENVIRONMENT *Georgi Markov, Atidzhe Ahmed*


P02_03 GROWTH LIGHT INTENSITY EFFECT ON THE STRUCTURAL STABILITY OF LHCII IN PEA THYLAKOIDS *Nia Petrova, Svetozar Stoichev, Svetla Todinova, Stefka Taneva, Sashka Krumova*

 **P02_04 COMMUNITY STRUCTURE AND INDIVIDUAL REACTION OF CHIRONOMIDAE (DIPTERA) LARVAE IN ANTHROPOGENICALLY IMPACTED SITES OF DANUBE RIVER (NE BULGARIA)** *Mila Ihtimanska, Julia Ilkova, Paraskeva Michailova*

P02_05 MORPHOGENESIS AND DEVELOPMENTAL PATTERNS AFFECT POLYPHENOLICS PRODUCTION AND ENDOGENOUS STRESS HORMONES IN *ARTEMISIA ALBA TURA* IN VITRO *Kalina Danova, Vaclav Motyka, Petre Dobrev*

 **P02_06 CHANGES OF ANTIOXIDATIVE ENZYMES DURING NATURAL REGENERATION IN DECIDUOUS FORESTS** *Vasil Chavgov, Detelina Petrova, Miroslava Zhiponova, Ganka Chaneva, Adriana Marinova, Svetoslav Anev, Nikolina Tzvetkova*

 **P02_07 DYNAMICS OF THE HEALTH STATUS OF COPPICE OAK FORESTS IN SOUTHWESTERN BULGARIA** *Nikolay Zafirov, Krasen Banov*

 **P02_08 IMPACTS OF THE FUNGICIDE AZOXYSTROBIN ON SOIL BACTERIAL COMMUNITIES** *Michaella Aleksova, Jordan Manasiev, Silvena Boteva, Mladen Popov, Anelia Kenarova, Galina Radeva*

P02_09 PROTEIN/RNA RATIO OF QUIESCENT AND EXPONENTIALLY GROWN CELLS OF SACCHAROMYCES CEREVISIAE *Anna Tomova, Anna Kujumdzieva, Ventsislava Petrova*

 **EP02_01 HIGH-LIGHT-INDUCIBLE SMALL STRESS PROTEINS AND THEIR LOCALIZATION IN THE CHLOROPHYLL-PROTEIN COMPLEXES OF CYANOBACTERIA** *Lubov Sharapova, Yuliya Oleskina, Nadezhda Yurina*

ECOLOGY AND EDUCATION

P05_01 INNOVATIVE INTEGRATED TRAINING IN HEALING PLANTS BUSINESS – TBP *Anna Kujumdzieva, Trayana Nedeva, Aleksander Savov, Tereza Toncheva*

P05_02 BLENDED LEARNING IN BIOINFORMATICS – THE SMES INSTRUMENT FOR BIOTECH INNOVATIONS – BIOTECH-GO *Anna Kujumdzieva, Trayana Nedeva, Aleksander Savov, Tereza Toncheva*

LANDSCAPE ECOLOGY

P04_01 RADIOLOGICAL MONITORING OF DRINKING WATERS IN BULGARIA *Radoslava Lazarova, Ivanka Yordanova, Donka Staneva*

OTHER RELATED TOPICS

 **P06_01 SUPER-CRITICAL CARBON DIOXIDE AS EFFECTIVE ECO-FRIENDLY SOLVENT FOR EXTRACTION OF PURSLANE (*PORTULACA OLERACEA* L.)** *Sabina Taneva, Svetlana Momchilova, Angel Konakchiev, Vladimir Dimitrov*

27 April 2018

**THEMATIC SESSION
BIODIVERSITY AND CONSERVATION BIOLOGY**

Chairmen: Prof. Boyko Georgiev and Prof. Vlada Peneva

Plenary presentations

9⁰⁰-9²⁰- BIOTECHNOLOGICAL APPROACH FOR PLANT PROPAGATION AND CONSERVATION IN IBER: AIMS AND ACHIEVEMENTS *Marina Stanilova-PL01_01*

9²⁰-9⁴⁰- WHO POLLINATES THE MEDICINAL PLANTS? *Ekaterina Kozuharova - PL01_02*

9⁴⁰-10⁰⁰- THE SECRETS OF PREDATORY NEMATODES (MONONCHIDA) *Vlada K. Peneva, Aleksandar Mladenov, Milka Elshishka, Stela Lazarova- PL01_03*

10⁰⁰-10²⁰ Discussion


10²⁰-10⁴⁰ - Coffee break

Oral presentations

10⁴⁰-10⁵⁰- THE ANTHROPOPHYTE FLORA OF LOZENSKA MOUNTAIN *Plamen Glogov, Dolja Pavlova- L01_01*


10⁵⁰-11⁰⁰- NEW INSIGHTS INTO MORPHOLOGY AND TAXONOMY OF THE PHOXINUS COMPLEX IN BULGARIA *Nina Bogutskaya, Tihomir Stefanov, Oleg Diripasko- L01_02*

11⁰⁰-11¹⁰-  MITOCHONDRIAL DIVERSITY OF BULGARIAN NATIVE DOGS SUGGESTS DUAL PHYLOGENETIC ORIGIN *Miroslav Marinov, Denitsa Teofanova, Georgi Radoslavov, Peter Hristov- L01_03*

11¹⁰-11²⁰-  NEW DATA ON MORPHOLOGY AND TAXONOMY OF THE FISHES FROM GENUS *ALBURNOIDES* (ACTINOPTERYGII, CYPRINIDAE) IN SOUTH-EASTERN BALKANS (AEGEAN AND BLACK SEA BASINS) *Tihomir Stefanov- L01_04*

11²⁰-11³⁰-  TOOLS OF PUBLIC RELATION IN HELP OF CONSERVATION BIOLOGY *Elisaveta Kozuharova, Danail Danov- L01_05*

11³⁰-11⁴⁰- STUDY ON MACROZOOBENTHIC TROPHIC STRUCTURE OF TEMPORARILY DRY RIVERS FROM BULGARIAN RIVER TYPE R14 *Rabia Soufi, Maria Kerakova, Emilia Varadinova L01_06*

11⁴⁰-11⁵⁰  ECOLOGICAL STATUS ASSESSMENT OF MOUNTAINOUS AND SEMI-MOUNTAINOUS STREAMS VIA DIFFERENT BIOTIC INDICES BASED ON BENTHIC MACROINVERTEBRATES - THE CASE STUDY OF MACEDONIAN-BULGARIAN CROSS-BORDER TERRITORY Biljana Rimcheska, Yanka Vidinova L01_07

11⁵⁰-12²⁰-Discussion

12²⁰-13²⁰-Lunch

Chairmen: Assoc. Prof. Ekaterina Kozuharova and Assoc. Prof. Ventsislav Karamfilov

13²⁰-13³⁰ - WHAT DO WE KNOW ABOUT RUDERAL FLORA AND VEGETATION IN BULGARIA? Kiril Vassilev, Mediya Gumus, Rayna Natcheva, Veselin V. Shivarov, Borislav Grigorov, Stoyan Georgiev, Nikolay Velev L01_08


13³⁰-13⁴⁰ - RELATIONSHIP BETWEEN GENETIC AND CHEMOTYPIC DIVERSITY IN PERENNIAL MEDICINAL PLANT *GLAUCIUM FLAVUM* CRANTZ (PAPAVERACEAE) Galya Petrova, Stefan Petrov, Iva Doycheva, Stoyan Stoyanov, Marina Stanilova L01_09

13⁴⁰-13⁵⁰ - GENUS *PSEPHELLUS* (ASTERACEAE) IN THE BULGARIAN FLORA Svetlana Bancheva, Malina Delcheva L01_10

13⁵⁰-14⁰⁰ -  NEW AND AMONG THE LARGEST POPULATIONS OF THE ENDANGERED ORCHID *GOODYERA REPENS* (L.) R. BROWN IN BULGARIA – STATUS Andrey Popatanasov L01_11

14⁰⁰-14¹⁰ -  FIRST RESULTS ON OF *SPERMOPHILUS CITELLUS* DISPERSAL AFTER REPATRIATION IN BULGARIA Maria Kachamakova, Yordan Koshev L01_12

14¹⁰-14²⁰ - A PHYTOCHEMICAL INSIGHT ON *CLINOPODIUM VULGARE* L. (LAMICEAE) Krum Bardarov, Reneta Gevrenova, Dimitrina Zheleva-Dimitrova and Ventsislav Bardarov L01_13

14²⁰-14³⁰ -  THE USE OF THE INDEX "% OLIGOCHAETA" IN DETERMINATION OF ECOLOGICAL STATUS AND POTENTIAL OF STANDING WATER BODIES Galia Georgieva, Yordan Uzunov L01_14

14³⁰-15⁰⁰ Discussion

15⁰⁰-15²⁰ - Coffee break

THEMATIC SESSION

ECOSYSTEM RESEARCH, SERVICES AND ECOLOGICAL AGRICULTURE

Chairmen: Prof. Valko Biserkov and Assoc. Prof. Stela Lazarova

Plenary presentations

15²⁰-15⁴⁰ MAPPING AND ASSESSMENT THE CONDITION AND ECOSYSTEM SERVICES OF INLAND WETLANDS IN BULGARIA OUTSIDE THE NATURA 2000 ECOLOGICAL NETWORK *Nevena Ivanova - PL03_01*

15⁴⁰-16⁰⁰ MAPPING OF ECOSYSTEMS TYPES, CONDITIONS AND THEIR SERVICES. CHALLENGES AND RECENT ACHIEVEMENTS IN THE BULGARIAN BLACK SEA REGION *Ventsislav Karamfilov, Dimitar Berov, Valko Biserkov, Georgi Daskalov, Stephania Klayn-PL03_02*

Oral presentations

16⁰⁰-16¹⁰  MACROPHYTES-BASED ECOLOGICAL STATUS ASSESSMENT OF DRAGOMANSKO AND ALDOMIROVSKO MARSHES *Teodora Ilieva, Gana Gecheva, Kiril Vassilev-L03_01*

16¹⁰-16³⁰ Discussion


16³⁰-17⁰⁰ - Coffee break

17⁰⁰-19⁰⁰ Poster sessions (BIODIVERSITY AND CONSERVATION BIOLOGY; ECOSYSTEM RESEARCH, SERVICES AND ECOLOGICAL AGRICULTURE) and Discussion

Chairmen: Assoc. Prof. Antoaneta Petrova and Assoc. Prof. Marina Stanilova

POSTER SESSION

BIODIVERSITY AND CONSERVATION BIOLOGY

 P01_01 RED DEER (*CERVUS ELAPHUS L.*) POPULATION DENSITY IN THE CENTRAL BALKAN MOUNTAINS (BULGARIA) REVEALED BY CAMERA TRAPS *Elitsa Popova, Ivan Stepanov, Atidzhe Ahmed, Peter Genov, Ivan Todev*

P01_02 MOLECULAR CHARACTERIZATION AND FIRST BULGARIAN RECORD OF *BUCHWALDOBOLETUS SPHAEROCEPHALUS* *Boris Assyov, Fuat Bozok, Hatıra Taşkın*

P01_03 PLANETELLA LIRONIS (ANTHRACOIDEACEAE) – A NEW RECORD FROM GREENLAND Teodor T. Denchev & Cvetomir M. Denchev


P01_04 PANGEON MT IN NORTH-EASTERN GREECE - NATURAL BOTANICAL GARDEN AND WORLD NATURE WEALTH Asen Asenov

P01_05 HYDROPONIC APPROACH FOR SEED PROPAGATION OF HABERLEA RHODOPENSIS FRIV. (GESNERIACEAE) Boryanka Traykova, Marina Stanilova

P01_06 INTRODUCTION OF IN VITRO CONDITIONS OF THE WILD MENTHA PULEGIUM L. Stanislava Stateva, Katya Uzundzhaliyeva

 **P01_07 BRYOPHYTES AND LARGER FUNGI ON THE TERRITORY OF VRANA PARK** Galina Gospodinova, Aneta Lambevska-Hristova, Rayna Natcheva, Melanya Gyosheva

P01_08 IMPACT OF SMALL HYDROPOWER PLANTS (SHPP) ON ECOLOGICAL STATE AND MACROZOOBENTHIC TROPHIC STRUCTURE (ISKAR RIVER, BULGARIA) Maria Kerakova, Stefan Kazakov, Emilia Varadinova, Luhezhar Pehlivanov

 **P01_09 DIVERSITY AND DISTRIBUTION OF BENTHIC DIATOM ASSEMBLAGES IN SMALL AND MEDIUM-SIZED BLACK SEA RIVERS AND ASSESSMENT OF THEIR ECOLOGICAL STATUS** Tsvetelina Isheva & Nadya Georgieva Ognyanova Rumenova

 **P01_10 WHAT DO WE KNOW ABOUT VEGETATION AND HABITAT DIVERSITY OF STRAZHATA HILL, NORTHERN BULGARIA?** Momchil Nazurov, Gana Gecheva, Constantin Mardari, Kiril Vassilev

P01_11 IN VITRO PROPAGATION OF ASTRACANTHA THRACICA (FABACEAE) Irena Mincheva, Marina Stanilova, Svetlana Bancheva, Malina Delcheva

P01_12 NEW RECORDS OF BULGARIAN HYPOGEOUS FUNGI Boris Assyov, Monica Slavova

P01_13 NEW DATA ON THE SPECIES OF ORCHIDACEAE IN THE STRANDZHA NATURAL PARK Svetla Dalakchieva, Antoaneta Petrova


P01_14 NEW ALIEN CONIFEROUS SPECIES IN BULGARIA Antoaneta Petrova, Rosen Vassilev, Kamen Bakardzhiev, Boris Assyov

P01_15 PARASITOIDS AND INQUILINES ASSOCIATED WITH GALLS OF ANDRICUS QUERCUSTOZAE (HYMENOPTERA: CYNIPIDAE) IN BULGARIA Ivaylo Todorov, Boyan Zlatkov, Anelia Stojanova, George Melika

 **P01_16 ETHNOBOTANY AND EXPLOITATION OF MEDICINAL PLANTS IN THE RHODOPE MOUNTAINS** Irena Mincheva, Michaela Jordanova, Ina Aneva, Ekaterina Kozuharova

P01_17 NEW SPECIES OF *CRISTAMPHIDELUS* SIDDIQI AND VINCIGUERRA, 1991 (NEMATODA: ALAIMIDA) FROM LIVINGSTON ISLAND, MARITIME ANTARCTIC *Milka Elshishka, Stela Lazarova, Georgi Radoslavov, Vlada K. Peneva*

ECOSYSTEM RESEARCH, SERVICES AND ECOLOGICAL AGRICULTURE

 **P03_01 IDENTIFICATION OF ECOSYSTEM TYPES IN HIGH MOUNTAIN AREAS: A CASE STUDY IN RILA MOUNTAIN** *Kostadin Katrandzhiev, Svetla Bratanova-Doncheva, Stoyan Nedkov*

P03_02 APHID PEST SPECIES ON *CHRYSANTHEMUM MORIFOLIUM* RAMAT. IN BULGARIA *Mariya Yovkova, Aneliya Pencheva*

P03_03 PRELIMINARY RESULTS OF THE EUTROPHICATION ASSESSMENT OF THE WATER BODIES IN THE LOWER DANUBE FLOODPLAIN USING REMOTE SENSING IMAGES AND *IN-SITU* VERIFICATION *Stefan Kazakov, Valko Biserkov, Luchezar Pehlivanov, Stoyan Nedkov*

 **P03_04 WEED SPECIES DIVERSITY AND COMMUNITY COMPOSITION IN ORGANIC POTATO FIELD** *Iliana Gerasimova, Totka Mitova*

19⁰⁰-19³⁰ Film

19³⁰ AWARDS AND CLOSING PROCEDURE OF THE 11th ANNUAL “SEMINAR OF ECOLOGY - 2018” WITH INTERNATIONAL PARTICIPATION

 **ABSTRACTS**
THEMATIC SESSION I
BIODIVERSITY AND CONSERVATION BIOLOGY

PL01_01

Biotechnological approach for plant propagation and conservation in IBER: aims and achievements

Marina Stanilova

Institute of Biodiversity and Ecosystem Research, Bulgarian Academy of Sciences

Aim: Plant biotechnologies englobe numerous methods, from rapid *in vitro* micropropagation to genome manipulation, oriented to creation and/or production of valuable or improved plants, and plant-derived products. Biotechnological Laboratory of Medicinal Plants, set up in 2001 at former Institute of Botany, later upgraded as part of Institute of Biodiversity and Ecosystem Research, introduced biotechnological approach toward conservation of endangered plant species from the Bulgarian flora, with special emphasis on medicinal plants production.

Material and methods: Seeds are used as most appropriate initial material for *in vitro* reproduction of plants of conservation importance, to preserve species genetic diversity. Seed germination was stimulated by different treatments. Besides, *in vitro* cultures were initiated from vegetative organs of previously selected individuals with valuable characteristics, to multiply genetically identical plants intended for agriculture. Plants were gradually *ex vitro* adapted in phytotron, then acclimated to open-air conditions.

Results: High-productive *in vitro* clones of *Leucojum aestivum* were selected, and *in vitro* galanthamine biosynthesis was studied in long-term cultures. *Ex situ* collections were established from *in vitro* plants of *Centaurea davidovii*, *C. pseudaxillaris*, *Alchemilla mollis*, *A. achtarowii*, *A. jumrukchalica*, *A. bundericensis*. Seed germination difficulties were overcome in recalcitrant species as *Astragalus physocalyx*, *Astracantha thracica*, *Glycyrrhiza glabra*. Plants of endemics were used to reinforce their original populations, among them *Verbascum tzar-borisii*, *V. davidoffii*, *V. anisophyllum*. Pilot plantation was established with high-productive *Valeriana officinalis* plants multiplied by clonal propagation.

Conclusion: Biotechnological approach proved to be successful for rapid multiplication of medicinal plants with resource deficiency, and for conservation of endemics.

Acknowledgments: Research was supported by several projects with international and national financing, among them: NATO SfP 974453, BG0034 EEA Grants/Norway grants, DFNI-BO2/18 Bulgarian National Science Fund.

Keywords: Green biotechnology, Medicinal plants, Bulgarian and Balkan endemics, Plant conservation, Agriculture

PL01_02

Who pollinates the medicinal plants?

Ekaterina Kozuharova

Medical University of Sofia, Faculty of Pharmacy, Department of Pharmacognosy, Sofia, Bulgaria

Medicinal plants are living creatures and we relate on their reproduction to supply the necessity of plant substances.

The **aim** of this study is to evaluate the necessity of pollinators for the reproduction of medicinal plants.

The plants recognized by Medicinal Plants Act (Bulgaria as a model) were classified as spore and gymnosperm plants, anemophylous (wind pollinated) plants and entomophylous (insect pollinated) plants. The plants that require insect vectors for their pollen transport are analyzed further. Entomophylous pollination syndromes are discussed according to the functional morphology and access to nectar and pollen of their “blossoms” (flower or compact inflorescence) in the following basic classes: “dish/bowl”, “bell”, “funnel” “flag” and “gullet”. Adaptations like spontaneous self-pollination ability and apomixis are also discussed.

The analysis roughly predicts the pollinators. About 85% of the medicinal plants depend on insects (bees predominantly) for their seed set.

What is the impact of pollinator decline on the resources of medicinal plants? Detailed research tests are needed for each particular plant, but expectations are for decline of the plant populations too.

Keywords: pollinators, bees, *Bombus*, medicinal plants

PL01_03

The secrets of predatory nematodes (Mononchida)

Vlada K. Peneva, Aleksandar Mladenov, Milka Elshishka, Stela Lazarova

Institute of Biodiversity and Ecosystem Research, Bulgarian Academy of Sciences

Predatory nematodes, especially mononchids, have attracted the attention of scientists since a very long time due to their peculiar morphology and trophic behaviour. The mononchid species *Prionchulus muscorum* (Dujardin, 1845) Bastian 1865 is among the first terrestrial nematodes described.

The **objective** of this work was to review the current state of research on predatory nematodes of order Mononchida

Material and methods: The review is based on literature data and original results.

Results: Currently, mononchids are represented by two suborders, 7 super-families and 556 species (Andrassy, 2009, Hodda, 2011). Being widely distributed in terrestrial and freshwater habitats, including Arctic and Antarctic, they represent different zoogeographic patterns. Common species have great variability and could represent genetically different species complexes, however molecular studies are yet insufficient. All representatives are predatory, prey consisting of protozoan, rotifers, small oligochaetes, enchytraeids, other nematodes, with cannibalism occurring. Occupying higher position in the soil micro food web they contribute to the regulating ecosystem services and could be used for their assessment and as indicators of soil quality.

Conclusion: Despite the long history of research of mononchids a number of questions regarding their diversity, evolution, phylogenetic and ecology should be further studied and clarified.

Acknowledgements: The present study was supported by the Program for Support of Young Researchers and PhD Students at the Bulgarian Academy of Sciences (Grant no. 17-6/2017), Bulgarian Academy of Sciences and ANIDIV 3.

Keywords: Taxonomy, morphology, ecology, biogeography

L01_01

The Anthropophyte flora of Lozenska Mountain

Plamen Glogov¹, Dolja Pavlova²

¹ Forest Research Institute, Department of Silviculture, Bulgarian Academy of Sciences 132, St. Kliment Ohridski Blvd.
BG-1756 Sofia, Bulgaria

² Sofia University, Faculty of Biology, Department of Botany, blvd. Dragan Tzankov 8, 1164 Sofia, Bulgaria

Aims: To analyze the taxonomical structure, origin and distribution of the anthropophytic element on the territory of Lozenska Mt. and to assess the degree of synanthropization of the flora.

Material and methods: The study was carried out in the period 2002-2017. During the field studies the transect method was applied for collection and identification of the plant material. In view of possible comparisons and calculations of the synanthropic indices the surveyed area was divided into 4 sub-regions.

Results: On the territory of the Lozenska Mt. 274 species and 8 subspecies from 196 genera and 57 families were found. The anthropophytes were predominantly therophytes. In phytogeographical terms the character of the anthropophytic flora could be described as European-Asiatic with a strong boreal and less sub-mediterranean influence. Four species of nature conservation status were identified among the anthropophytes. The antropophytes of Lozenska Mt. were 33.3% of its vascular flora and almost twice of the same group of plants in the flora of Bulgaria (14%).

Conclusion: The north-eastern part of the mountain is most affected by the synanthropic processes and less affected is the north-western part. For the period 1961-2017 the percentage of the anthropophytes in the surveyed area is increased by 7.2%, while the participation of the autochtonous species is decreased by 3.8%. The high values of calculated indices of synanthropization show the intensification of this process on the territory of the mountain.

Keywords: synanthropization, apophytes, autochtonous flora, ecological groups

L01_02

New insights into morphology and taxonomy of the *Phoxinus* complex in Bulgaria

Nina Bogutskaya¹, Tihomir Stefanov², Oleg Diripasko³

¹Vienna Museum of Natural History, Burgring 7, 1010 Vienna, Austria

²National Museum of Natural History, 1 Tsar Osvoboditel Blvd, 1000 Sofia, Bulgaria

³Institute of Fisheries and Marine Ecology, 8 Konsulska St, Berdyansk, 71118, Ukraine

Recent publications revealed considerable molecular diversity of European *Phoxinus* only partly congruent with existing taxonomic concepts, and a high number of clades that lack clear morphological diagnosability.

Aim: The present study is devoted to morphology of *Phoxinus* in Bulgaria with special emphasis on meristic characters including age-and-size variability, geographic variation, and sexual dimorphism within and between putative species.

Material and methods: There were 235 examined specimens from five localities (three different basins) in Bulgaria and 520 specimens from adjacent regions for comparison. The examined characters include the pattern of scalation and the lateral-line canals and sensory canal structure, and the axial skeleton morphology (vertebral counts by skeletal regions and the number of precaudal anal-fin pterygiophores).

Results: The characters were used to demonstrate differences between two presumably distinct species: the Danubian *P. csikii*, a species recently resurrected based on molecular data, and *P. strandjae*. The redescription of *P. strymonicus* revealed its close morphological affinity to *P. strandjae*.

Conclusion: The pattern of morphological variation partly corresponds to the published molecular schemes and supports taxonomic validity of *P. csikii*, and *P. strandjae* in Bulgaria.

Acknowledgements: NB is supported by the FWF (Meitner Programme) project M 2183-B25.

Keywords: *Phoxinus*, morphology, taxonomy, variations, osteology, Bulgaria

L01_03

Mitochondrial diversity of Bulgarian native dogs suggests dual phylogenetic origin

Miroslav Marinov¹, Denitsa Teofanova², Georgi Radoslavov¹, Peter Hristov¹

¹Institute of Biodiversity and Ecosystem Research, Bulgarian Academy of Sciences, Department of Animal Diversity and Resources

²Sofia University, Faculty of Biology, Department of Biochemistry

The dog has been the first domesticated animal to have a central role in human society from ancient times to present day. Although there have been numerous investigations of dog phylogeny and origin, genetic data of dogs in the region of the Balkan Peninsula (South-Eastern Europe) are still scarce.

Therefore, the **aim** of the present study is to perform phylogenetic analysis of three native Bulgarian dog breeds.

Material and methods: A total of 130 samples were analyzed according to HVR1 (hypervariable region, D-loop region). The samples were taken from two hunting dog breeds (Bulgarian Hound dog: Barak, n=34; Bulgarian Scenthound dog: Gonche, n=45) as well as from a Bulgarian Shepherd dog (n=51). The first two breeds are reared in a flat region of the country (the Northern part of Bulgaria, the Danubian Plain), while the last breed is a typical representative of the mountainous part of the country.

Results: The results have shown the presence of almost all main clades – A, B, C and D – in the three dog breeds taken together, except clades E and F, as expected. With regard to haplogroups distribution, there are clear differences among investigated breeds. While hunting breeds exhibit a prevalence of the C clade, the mountainous Shepherd dog shows presence of the D2 haplogroup but absence of the C clade.

Conclusion: In conclusion, the present study is the first to investigate the mitochondrial diversity of native dog breeds in Bulgaria. The results show a clear difference of haplogroups dissemination in native hunting and shepherd dogs that suggests a dual independent phylogenetic origin without hybridization events between them.

Keywords: Bulgarian native dog, D-loop region, genetic diversity, population structure

L01_04

New data on morphology and taxonomy of the fishes from genus *Alburnoides* (Actinopterygii, Cyprinidae) in South-eastern Balkans (Aegean and Black Sea basins)

Tihomir Stefanov

National Museum of Natural History, Bulgarian Academy of Sciences

It has been considered for long time that genus *Alburnoides* in Europe and parts of Asia is represented by only one species – *Alburnoides bipunctatus*. Recent molecular study gives new light about the taxonomy and distribution of these fish in Europe and reveals 17 different Eurasian lineages. Four of them inhabit the territory of the South-eastern Balkans and need detailed morphological description.

Aims: The aim of the paper is to study the morphology of the main *Alburnoides* populations in the rivers flowing into Aegean and Black Seas and situated on the territory of the South-eastern Balkans. Based on this data particular taxonomic implications were made.

Material and methods: A total of 182 specimens from eleven localities were examined. The localities were situated on eight different river basins on the territory of three countries – Greece, Bulgaria and Turkey. A total of 22 morphometric and 7 meristic characters were used in the study.

Results: The study demonstrates differences in the morphology of the four different molecular lineages and describes them well as a distinct species.

Conclusion: The pattern of morphological variation well corresponds to the published molecular lineages and supports the taxonomic validity of the species *A. economoui*, *A. thessalicus*, *A. strymonicus* and *A. tzanevi*.

Keywords: *Alburnoides*, morphology, taxonomy, Balkan Peninsula

L01_05

Tools of Public Relations in help of conservation biology

Elisaveta Kozuharova, Danail Danov

Sofia University, Faculty of Preschool and Primary School Education, Department of Media Pedagogy

Aim: Often business interests and local people livelihood are in conflict with biodiversity conservation e. g. wind power stations at Kaliakra, Pirin Mts and ski runs, Kresna Gorge and highway. The aim of this work is to demonstrate how the tools of Public Relations (PR) can serve to the conservation biology.

Material and methods: The tools of Public Relations (PR) are as follow: 1) for effective results the approach of PR is to define adequately the problem; 2) next step is to choose the right type of analysis amongst: organizational or situational with their verities; 3) then based on the analyses' result a strategy is built.

Results: Here we present the basic strategic models. An important point is to keep track of the influence and feedback all the time. A possible example is presented devoted to the problem of bee decline.

Conclusion: The proper application of the tools of PR can effectively improve the protection of biodiversity.

Keywords: tools of Public Relations (PR), analysis, basic strategic models, conservation biology

L01_06

Study on macrozoobenthic trophic structure of temporarily dry rivers from Bulgarian river type R14

Rabia Soufi, Maria Kerakova, Emilia Varadinova

Institute of Biodiversity and Ecosystem Research, Bulgarian Academy of Sciences

Aim: The aim of this study is to analyze the macrozoobenthic trophic structure of the specific conditions of temporarily drying rivers (R14) situated in Southern Bulgaria, by analyzing the quantitative and qualitative composition of the functional feeding groups.

Material and methods: Macrozoobenthic samples from 25 river sites were collected during the two-year observation period (May 2014 and June 2015), like 8 of them being sampled and tested during both studying years. An adapted version of multi-habitat sampling method was used. Benthic taxa were determined and assigned to six functional feeding groups.

Results: The group of primary consumer – scrapers, was dominant (with nearly 50%) in the trophic structure of the temporarily drying rivers (R14). The other functional groups, presented as a mean value of the total number of individuals, were relatively equally represented and their relative share of the trophic structure was between 6% and 17%. In regard to completeness and qualitative composition of the trophic structure, as well as a relative share of the six functional groups, we found a radically different structure at the same river point during the two study years.

Conclusion: The specific conditions of the environment into drying rivers are a prerequisite for the formation of a distinct trophic structure, dominated by the scrapers, whose composition is prevailed by more tolerant taxa. Local effects of the anthropogenic pressure, drought regime and the period of recovery of the benthic community, build up differently at the same point during the two observed years, functional feeding groups' distribution.

Keywords: macrozoobenthos, functional feeding groups, trophic structure, scrapers

L01_07

Ecological status assessment of mountainous and semi-mountainous streams via different biotic indices based on benthic macroinvertebrates - the case study of Macedonian-Bulgarian cross-border territory

Biljana Rimcheska, Yanka Vidinova

Institute of Biodiversity and Ecosystem Research, Bulgarian Academy of Sciences,
1 Tsar Osvoboditel Blvd., 1000-Sofia, Bulgaria

Aim: The aim of this study is to provide indicative ecological status assessment of mountainous and semi-mountainous streams of a Macedonian-Bulgarian cross-border territory, based on benthic macroinvertebrates indices according to the requirements of the EU Water Framework Directive (WFD).

Material and methods: The sampling for this study was conducted in October 2017 at 10 sites. The bottom macroinvertebrate specimens were collected with hydrobiological hand net (mesh size 500 µm) applying kick&sweep multihabitat procedure. For indicative ecological status assessment the following metrics were used: BWMP, ASPT, IBI and EPT.

Results: The water quality of the examined sites was assessed from excellent to moderate (I - III class). Predicting that presence of any kind of habitat degradation or pollutants at any level causes observable changes over benthic composition in the studied sites, a comparison between the affected and clear sites was done. In addition, the taxonomic composition is examined in order to select indicator taxa among the endemic ones as they present in the studied area.

Conclusion: Our results contributed to the knowledge on the current ecological state of some rivers in a poorly studied area and assisted the selection of appropriate metrics for water quality assessment based on macroinvertebrates as BQE.

Acknowledgements: The study was supported by project №DFNP-17-108/28.07.2017 “Implementation of biotic indices BMWP and ASPT in order to evaluate the ecological status of mountain and semi-mountain rivers from the 7th Ecoregion (Eastern Balkans)”, funded by Bulgarian Academy of Science.

Keywords: Macrozoobenthos, Mountainous and semi-mountainous streams, BMWP, ASPT, IBI, EPT

L01_08

What do we know about ruderal flora and vegetation in Bulgaria?

Kiril Vassilev¹, Mediya Gumus², Rayna Natcheva¹, Veselin V. Shivarov¹, Borislav Grigorov³, Stoyan Georgiev⁴, Nikolay Velev¹

¹Institute of Biodiversity and Ecosystem Research, Bulgarian Academy of Sciences, Department of Plant and Fungal Diversity and Resources, Sofia, Bulgaria

²University of Plovdiv “Paisii Hilendarski”, Faculty of Biology, Plovdiv, Bulgaria

³University of Sofia “St. Kliment Ohridski”, Faculty of Geology and Geography, Sofia, Bulgaria

⁴Field Crops Institute - Chirpan, Agricultural Academy, Chirpan, Bulgaria

Aim: The aim of this study is to summarize the available data about syntaxonomical diversity of synantropic vegetation in Bulgaria.

Material and methods: The study area covers the whole territory of the country. We collected and analyzed all available data about synantropic vegetation from phytocoenological literature and databases – Balkan Dry Grassland Database and Balkan Vegetation Database. The high rank syntaxa follows the concept of Mucina et al. (2016). All localities were georeferenced by Google Earth *a posteriori*, in case of missing GPS data. Mapping was done using ArcGIS 10.0 software.

Results: We prepared groups of vascular plants, mosses and lichens, which distribution optimum is related to the synantropic vegetation. Recent knowledge on ruderal flora, including mosses and lichens, in Bulgaria is quite limited. Until now, the ruderal vegetation in the country is represented by 5 classes, 11 orders, 21 alliances, 42 associations and 8 plant communities without rank.

Conclusion: The data about ruderal flora, including mosses and lichens, and vegetation from the country were sporadic so far. We plan to reveal much more of the existing vegetation and floristic diversity in the country during the period 2018-2021, when many new relevés will be collected within the project “Study of ruderal flora and vegetation on the territory of Bulgaria and Romania”.

Keywords: anthropophytes, disturbed habitats, segetal flora, synantropic vegetation, weeds

L01_09

Relationship between genetic and chemotypic diversity in perennial medicinal plant *Glaucium flavum* Crantz (Papaveraceae)

Galya Petrova¹, Stefan Petrov², Iva Doycheva¹, Stoyan Stoyanov¹, Marina Stanilova¹

¹Institute of Biodiversity and Ecosystem Research, Bulgarian Academy of Sciences

²Institute of Molecular Biology “Roumen Tsanev”, Bulgarian Academy of Sciences

Aim: The current study investigates the genetic diversity of five Bulgarian populations of the medicinal plant species *Glaucium flavum* Crantz (Papaveraceae) classified into different chemotypes on the basis of our previous research.

Material and methods: Fresh leaves were collected from five populations along the Bulgarian Black Sea coast near the settlements of Shabla, Shkorpilovtsi, Pomorie, Varvara, and Sinemorets. Monitoring of populations was carried out according to the methodology approved by the national authorities. Total genomic DNA was isolated by modified CTAB method. The quantity and quality of DNA were estimated spectrophotometrically. A subset of samples was checked using agarose gel electrophoresis. Genetic diversity was assessed based on Inter Simple Sequence Repeat (ISSR) markers. All the results were tested for reproducibility. Allele frequency data were used to estimate summary diversity indices for each population using GenAlEx (v.6.5).

Results: Successful and reproducible amplifications were obtained based on selected primers. The smallest population (near Shabla) from the most spread glaucine profile exhibited the lowest level of genetic diversity, while the only population belonging to isocorydine chemotype (near Shkorpilovtsi) was characterised by the uppermost level of genetic diversity. The overall differentiation among populations was very high.

Conclusion: The molecular analysis, together with our field observations indicate that all populations require regular monitoring and well-designed conservation measures. The population, exhibiting the lowest genetic diversity and characterized by its richness in glaucine should be prioritised for conservation. More specific studies are necessary to explore the epigenetic basis of *G. flavum* chemotypes.

Acknowledgements: This research was supported by the Bulgarian National Science Fund (project # DFNI-B02/18).

Keywords: yellow hornpoppy, genetic diversity, chemotypes, monitoring, conservation

L01_10

Genus *Psephellus* (Asteraceae) in the Bulgarian flora

Svetlana Bancheva, Malina Delcheva

Institute of Biodiversity and Ecosystem Research, Bulgarian Academy of Sciences, Acad. G. Bonchev, bl. 23, Sofia, Bulgaria

Genus *Psephellus* Cass. (Asteraceae) is represented in the Bulgarian flora by two species, *P. marschallianus* (Spreng.) C. Koch and *P. trinervius* (Willd.) Wagenitz, both of which are very rare and are included in the Bulgarian Biodiversity Conservation Act. In the Bulgarian literature to date, these two species have been included within the range of the genus *Centaurea* L.

Aim: This study aims to clarify the correct taxonomic status of the representatives of genus *Psephellus* in Bulgaria, to assess their population state and to comment the needs and measures for their conservation.

Material and methods: The investigation is carried out using all available literature data and herbarium specimens stored in the Bulgarian Herbaria, as well as the personal collections and field observation on the species' populations in Bulgaria. Conservation measures are undertaken in the unique Bulgarian population of *P. trinervius* on Taushan tepe locality.

Results: *P. marschallianus* is currently known only from several localities in two floristic regions of Bulgaria, Black Sea Coast (Northern) and North East Bulgaria. Populations are with 100–200 individuals each, in good state, but strongly fragmented. *P. trinervius* is known from unique population with several individuals in North East Bulgaria floristic region. It grows in dry sunny places with sparse vegetation on clayey marls. An action plan has been developed to preserve the species. Appropriate *in situ* measures have also been taken.

Conclusion: It is expected that the conservation status of the populations of the target species will be improved.

Acknowledgments: Financial support by the National Fund “Scientific Research” of Bulgaria, under Grant DN 01/7-16.01.2017 ‘Flora of the Republic of Bulgaria, vol. 12: Biological Diversity in Asteraceae subfam. *Carduoideae* and *Cichorioideae*’ is gratefully acknowledged.

Keywords: Asteraceae, Plant conservation, *Psephellus*

L01_11

New and among the largest populations of the endangered orchid *Goodyera repens* (L.) R. Brown in Bulgaria – status

Andrey Popatanasov

Faculty of Biology, Plovdiv University “Paisij Hilendarski”, Plovdiv, Bulgaria

Aim: *Goodyera repens* (L.) R. Brown is small inconspicuous relict boreal orchid found only in century old forests. Due to its peculiar biology and environmental requirements it's among the most threatened vascular plants in Bulgaria. It fulfills the IUCN criteria for endangered [EN B1ab(iii)+2ab(iii)] therefore included in Red Data Book and Biodiversity Act. The study aimed to explore and evaluate its distribution and bioconservatory status in Trigrad- Yagodina plateau.

Materials and methods: Exploration and monitoring of *G. repens* populations in Trigrad- Yagodina karst plateau was done from 2012 until 2017. Shoot count and GPS coordinates caching were performed for distribution mapping.

Results: During the monitored period six new locations of *G. repens* for Bulgaria were found on area of several square kilometers in the coniferous belt between the villages Trigrad and Yagodina. At each location shoot number ranged from few tens to several hundred leaf rosettes. However by the end of monitored period four locations were destroyed by wood cutting and forest clearing activities. At the fifth location over 60% of shoots and occupied area were destroyed. Both of the largest survived locations were in suppressed state due to anthropogenic factors.

Conclusion: Bulgaria is the southernmost border of the species global distribution areal which puts high stress on population survival and distribution. Although there's high count at some of the locations the population's genetic diversity is rather poor due to dominantly vegetative reproduction. These facts necessitate restriction of anthropogenic activities in these habitats and give some of them a protected status.

Keywords: Creeping Lady's Tresses, Bioconservation, Orchidaceae

L01_12

First results on of *Spermophilus citellus* dispersal after repatriation in Bulgaria

Maria Kachamakova and Yordan Koshev

Institute of Biodiversity and Ecosystem Research, Bulgarian Academy of Sciences

Aim: During the last decades repatriation activities have been widely implemented to improve European ground squirrel [EGS] (*Spermophilus citellus*) conservation status, but until now no detailed study of the post-release spatial behavior has been implemented in South-Eastern Europe. That is why we consider the inclusion of radiotelemetry in new reinforcement action in “Zapadna Strandzha” SPA, Bulgaria, as precious opportunity to provide data about the process of adaptation and dispersal.

Material and methods: 18 of 96 repatriated animals were equipped with radio-collars. Radio-telemetry was performed for 13 days in total from July to September. At the end of each day the location of the animal (the burrow currently inhabited) was noted. For each location the distance to the mean release point was calculated. Analysis of covariance (ANCOVA) was performed to investigate the variation of the mean and the maximal distance travelled among the sexes and the ages.

Results: We found that the male individuals undertake significantly longer dispersal than the females based on both mean (for males 217±50 m, for females 91±20 m; n=14, p<0.05) and maximal (for males 252±42 m; for females 130±23 m; n=14, p<0.05) distance travelled. The age has marginal positive influence (p<0.1). There were no evidences for interaction between age and sex factors.

Conclusion: These preliminary results show that the widely observed pattern that male ESG show increased boldness and dispersal stays valid also in totally new environment and after an artificial intervention. It should be taken into account while planning such conservation activities in future.

Acknowledgments: The reinforcement and radio telemetry study were implemented and presented under the LAND for LIFE project (LIFE14 NAT/BG/001119, coordinated by the Bulgarian Society for the Protection of Birds and co-financed by the EC LIFE Programme) and the Bulgarian Academy of Sciences Young Scientist Grant № DFNP-17-110.

Keywords: reinforcement, dispersal, conservation biology, ground squirrel

L01_13

A phytochemical insight on *Clinopodium vulgare* L. (Lamiceae)

Krum Bardarov¹, Reneta Gevrenova², Dimitrina Zheleva-Dimitrova² and Ventzislav Bardarov³

¹ Institute of molecular biology and biotechnology, boul. Riski 105, Plovdiv-4000, Bulgaria

² Department of Pharmacology, Pharmacotherapy and Toxicology, and Pharmacognosy, Faculty of Pharmacy, Medical University of Sofia, Dunav 2 str., Sofia-1000, Bulgaria

³ Chromana Ltd, Sampkov 78 str., Sofia-1113, Bulgaria

Ethnopharmacological relevance: *Clinopodium vulgare* L. (Lamiceae) is a traditional plant in Bulgarian folk medicine for treatment of diabetes, gastric ulcers and cancer. The aerial parts alleviate symptoms associated with mastitis and prostatitis. A recent *in vivo* study showed strong antioxidant capacity, discerned by statistically significant increased activities of the antioxidant enzymes and decreased production of malondialdehyde.

Aim of the study: To analyze the lyophilized water extract by ultra high performance liquid chromatography (UHPLC) combined with high resolution mass spectrometry (HRMS).

Material and methods: The lyophilisate of the hot water extract from the dried aerial parts of *C. vulgare* was analyzed by UHPLC-HRMS using a quaternary pump and a hybrid quadrupol-Orbitrap high resolution Q-Exactive mass spectrometer coupled with a heated electrospray ionization (HESI) probe. The chromatographic separation was performed on Poroshell C18 3 x 150 mm 2.7 um column with gradient of 10÷95% acetonitrile in 0.08% formic acid as mobile phase at flow rate 250 ul/min, and on Silica-HILIC 150 x 3 mm 2.6 um column with 5÷50% gradient of water in acetonitrile with 10 mM ammonium formate pH 4.6 at flow rate 450 ml/min.

Results: Based on the HRMS and MS/MS data, as well as comparison with reference substances and literature data, more than twenty secondary metabolites were identified or tentatively elucidated in *C. vulgare* extract. A variety of phenolic (ferulic, coumaric) and mono- and di-acylquinic acids were evidenced together with clinopodic acids B, C, E, G, H, |K and their isobars. Flavonoids belong to flavons, flavonols and flavonons, being presented by O- and C-glycosides. Among the major compounds were rosmarinic acid, clinopodic acid K and Clinoposaponin I.

Conclusion: The results highlighted *C. vulgare* as a valuable source of bioactive compounds, extracted with water in more than 20% from dried aerial parts of the plant.

Keywords: *Clinopodium vulgare*, UHPLC, HRMS, MS/MS

L01_14

The use of the index "% Oligochaeta" in determination of ecological status and potential of standing water bodies

Galia Georgieva, Yordan Uzunov

Institute of Biodiversity and Ecosystem Research, Bulgarian Academy of Sciences, Department of Aquatic Ecosystems

Aim: The present study is aiming at implementing the metric "% Oligochaeta" for assessment of the ecological status, potential and environmental classification of standing water bodies.

Material and methods: The survey was based on published and still unpublished data. Samples were taken during 2003; 2007; 2011; 2012; 2013 and 2017 in the littoral area of standing water bodies in Bulgaria. Studied sites referred to the eco-regions Eastern Balkan and Pontic Province, and belong to different types of stagnant water bodies, according to the adopted national typology of freshwater bodies (Ordinance No 4/2013).

Qualitative samples of benthic macroinvertebrates were taken according to the standard ISO/EN/BDS methods. An adapted version of the multi-habitat sampling method was applied. The species composition and abundance of the macrozoobenthos were measured and further "% Oligochaeta" was calculated.

Results: The index was designed to characterize the abundance of the macrozoobenthic fauna in lakes and reservoirs and it was experimentally tested and standardized for the purpose of developing a classification system for assessment of the ecological status/ potential of different types of surface water bodies. The Oligochaeta fauna of the studied stagnant water bodies, both lakes and reservoirs, was composed of tolerant species, easily adaptive to different environmental conditions including anthropogenic impacts.

Conclusion: The index has great potential for hydrobiological assessments, but the vast typological diversity of standing water bodies in our country, that include artificial and highly modified surface water bodies, and their intensive economic use, does not give enough reliable statistical confirmations so further research is needed to build up more data and conclusions.

Keywords: lakes, %Oligochaeta index, abundance, water quality

P01_01

Red deer (*Cervus elaphus* L.) population density in the Central Balkan Mountains (Bulgaria) revealed by camera traps

Elitsa Popova¹, Ivan Stepanov², Atidzhe Ahmed³, Peter Genov³, Ivan Todev⁴

¹Sofia University "St. Kliment Ohridski", Faculty of Biology, Blvd. "Dragan Tsankov" 8, Sofia 1164

²State Hunting Enterprise "Rositsa", Stokite locality

³Institute of biodiversity and ecosystem research, BAS, Blvd. "Tsar Osvoboditel" 1, Sofia 1000

⁴Southwestern State Enterprise, Blagoevgrad, str. "Zora"18

Aim: The main aim of the presented study is to apply the random encounter model (REM) for estimation of the population density of red deer in a mountainous hunting area based on camera trap data. This method has been proved to provide accurate results for ungulates when sufficient field effort and positioning of the camera traps is applied. Furthermore, it can provide a scientifically sound method for game census, which would replace the old and inaccurate methods currently used.

Material and methods: 38 camera traps were set up in the State Hinting Enterprise "Rositsa", on the Northern slopes of the Central Balkan Mountains, Bulgaria in July 2018 at a distance of approx. 1 km from each other. A total of 702 camera trap days were accumulated. The resulting photographs and videos were analyzed through Camera Base and the random encounter model (REM).

Results: The results indicate a population density of 4.72 ind./km², which is much lower than the reported by the official census. This value is above the average for red deer populations studied in other parts of Europe. The Hunting enterprise provides supplementary feeding in the winter months, as well as protection against poaching, which contributes to the wellbeing of the population.

Conclusion: The applied method of estimating population density based on camera trap data is cost-efficient, requires limited field effort and provides scientifically sound results. It could be widely used to monitor mammal populations, especially game species such as the red deer, where the hunting quotas are based on census data.

Acknowledgments: This work was supported by project № 195/10.10.2016 “Population studies, migration and status of the red deer (*Cervus elaphus*) in Bulgaria” of the Southwestern State Enterprise, Blagoevgrad, and the Institute of biodiversity and ecosystem research, BAS. We thank the staff of the State Hunting Enterprise for their cooperation and contribution.

Keywords: Red deer, camera traps, random encounter model

P01_02

Molecular characterization and first Bulgarian record of *Buchwaldoboletus sphaerocephalus*

Boris Assyov¹, Fuat Bozok², Hatıra Taşkın³

¹Institute of Biodiversity and Ecosystem Research, Bulgarian Academy of Sciences, Acad. G. Bonchev st. 23, 1113 Sofia, Bulgaria

²Osmaniye Korkut Ata University, 8000 Osmaniye, Turkey

³Çukurova University, 01330 Adana, Turkey

Aim: The aim of this work is to expand the knowledge on boletes, providing ITS characterization of *Buchwaldoboletus sphaerocephalus* on collection from South-eastern Europe.

Material and methods: Specimens were characterized macromorphologically and microscopically. DNA was isolated by the procedures of Eurx Genematrix Plant & Fungi DNA Purification Kit. ITS region was amplified by ITS1F-ITS4 primers. Sequencing was made with ABI 3730XL Sanger sequencer. The analysis was run on the phylogeny.fr platform.

Results: Few sequences of the genus *Buchwaldoboletus*, mostly unpublished, are available on public databases. In this study we obtained ITS sequence of the first Bulgarian collection of *B. sphaerocephalus*. The analysis revealed that it resolves in a well-supported clade with two sequences labelled as *B. hemichrysus* from USA and UK. The Bulgarian collection is further characterized morphologically and discussed.

Conclusion: The genus *Buchwaldoboletus*, with its 13 taxa worldwide is currently underrepresented by sequences and the number of species and their relationships are unresolved by phylogenetic methods. The high similarity between European and North American sequences supports the unsettled conspecificity between *B. hemichrysus* and *B. sphaerocephalus*, but analysis of more extensive set of materials, including types and topotypic specimens, is still necessary.

Acknowledgements: Studies of the first author are within project ‘Taxonomy, conservation and sustainable use of fungi’.

Keywords: *Balkan mycota*, Boletaceae, Boletales, phylogeny

P01_03

Planetella lironis (Anthracoideaceae) – a new record from Greenland

Teodor T. Denchev & Cvetomir M. Denchev

Institute of Biodiversity and Ecosystem Research, Bulgarian Academy of Sciences, Acad. G. Bonchev st. 23, 1113 Sofia, Bulgaria

Aim: a contribution to the taxonomy and distribution of the smut fungi in North America.

Material and methods: A dried specimen from the herbarium of University of Copenhagen (C) was examined under light microscope (LM) and scanning electron microscope (SEM). For LM observations and measurements, spores were mounted in lactoglycerol solution (w : la : gl = 1 : 1 : 2) on glass slides, gently heated to boiling point to rehydrate the spores, and then cooled. For SEM, spores were attached to specimen holders by double-sided adhesive tape and coated with platinum in an ion sputter. The surface structure of spores was observed and photographed at 10 kV accelerating voltage using a JEOL JSM 6610-LV scanning electron microscope.

Results and conclusion: *Planetella lironis*, a rare smut fungus known only from Canada, is reported for the first time from Greenland, on *Carex maritima*.

Acknowledgements: This study was funded by the Program for Support of Young Researchers and PhD Students at the Bulgarian Academy of Sciences (Grant no. DFNP-17-93/28.07.2017). The assistance of the staff of the herbarium of University of Copenhagen (C) is kindly acknowledged.

Keywords: *Anthracoideaceae*, *Carex*, Greenland, *Planetella*, smut fungi, taxonomy

P1_04

Pangeon Mt in North-Eastern Greece - natural botanical garden and world nature wealth

Asen Asenov

Sofia University, Faculty of Biology, Department of Botany

Aim: Primary investigation of vascular flora of Pangeon Mt.

Material and methods: Field trips

Introduction: Pangeon Mt (1956 m) is situated on North Eastern Greece, on the north coast of the Aegean Sea. Basic rocks are predominantly calcareous (marbles) and soils are predominantly rendzinas. Calcareous terrain is reason for especially rich flora. The climate is Transitional Mediterranean. The next plant belts are presented from the foothills to the top. Maquis, *Quercus* belt (*Quercetum mixtum*), beech belt and grassy subalpine belt. Typical subalpine grassy vegetation is found in the high belt, above 1500-1700 m.

Results: The old beech and relict forests of *Ostrya carpinifolia* as well as many Balkan endemic and relict plants as *Abies borisi-regis* Mattf., *Achillea ageratifolia* (Sm.) Boiss., *Anthyllis aurea* Welden, *Fritillaria drenovskyi* Degen & Stoj., *Gymnadenia conopsea* (L.) R. Br., *Haberlea rhodopensis* Friv., *Ostrya carpinifolia* Scop., *Petkovia orphanidea* (Boiss.) Stef., *Saxifraga ferdinandi-coburgi* Kellerer & Sünd., *S. sempervivum* C. Koch., *Sideritis scardica* Griseb., *Viola delphinantha* Boiss., *V. perinensis* Becker etc. is part of the vascular flora of Pangeon Mt.

Conclusion: Occurrence of endemics, relicts and rare species, some of them with narrow ecological niche is an indication of the great conservation importance of Mt Pangeon. Pangeon Mt is a treasury of world wealth.

Keywords: *Calcareous terrain*, endemic plants, vascular flora

P01_05

Hydroponic approach for seed propagation of *Haberlea rhodopensis* Friv. (Gesneriaceae)

Boryanka Traykova, Marina Stanilova

Institute of Biodiversity and Ecosystem Research, Bulgarian Academy of Sciences

Aim: The study is aiming to test the opportunities for propagation of the tertiary relict, Balkan endemic and medicinal plant *Haberlea rhodopensis* from seeds, using a hydroponic technique.

Material and methods: Seed-vessels of ten *H. rhodopensis* plants were gathered from the population in the Rhodopes, near Sitovo village, Plovdiv district, in October 2017. Dust-like seeds were germinated on Cutting Board hydroponic system comprising 27 pots with peat cubes surrounded with agrolava pebbles, dipped in standard nutrient solution (pH 5.5-6.5; EC 0.4-0.6 mS), at 20±4°C, air humidity 30-65%; darkened for a month, with mixed daily and artificial light 16/8 h after the germination start. In addition, seeds were put on 10 damp coco-fiber cubes, in a tray with transparent cover.

Results: First seeds germinated two weeks after their setting on the hydroponic system. At the end of the first month, numerous seeds germinated and formed small cotyledons on 23 pots. Seedlings growth was very low: rosettes with 5 mm diameter, consisting of 4-6 leaves, formed at the end of the third month. Rosettes at the pots periphery had more space and grew faster. Meantime, mosses developed on the peat cube surface and the substrate was infested by insects and fungi, causing putrefaction of some rosettes. Plantlets were treated with biofungicide and bioinsecticide. Largest 30 rosettes were transferred to tray with soil mixture and perlite (1:1). For comparison, few seeds on coco-fiber cubes germinated but began to necrotize.

Conclusion: First trials on *Haberlea rhodopensis* propagation from seeds using hydroponics are promising.

Acknowledgments: Authors are grateful to the Bulgarian Academy of Sciences, Program supporting young scientists and PhD-students (Contract # DFNP-17-96 / 28.07.2017). Authors highly appreciate the advices of Rastena EOOD.

Keywords: Orpheus flower, resurrection plants, seed germination, plant propagation, hydroponic systems

P01_06

Introduction of *in vitro* conditions of the wild *Mentha pulegium* L.

Stanislava Stateva, Katya Uzundzhaliyeva

Institute of Plant Genetic Resources - Sadovo, Plovdiv, Bulgaria

The **aim** of this study is to introduce *Mentha pulegium* L. *in vitro*.

Material and methods: The experimental work was carried out in 2017 in the laboratory of tissue culture at IPGR "Konstantin Malkov" - Sadovo. The fresh plant material was taken during an expedition along the Stryama River. Sterilization of explants of 1.0 - 3.0 cm length was performed with 0.3% HgCl₂ solution for 1, 2 and 3 min, followed by immersion in 98% ethyl alcohol for 3 s. Thus, the sterilized explants are cultured on culture medium - Murashige & Skoog (1962) without added hormones. Sucrose (30 g/l) was used as the carbohydrate source, and agar (7.0 g / l) hardener with a pH of medium 5.6. Exploitation was carried out in a temperature-controlled chamber of 22-25 ° C and 16 hours of darkroom photoperiod and 8 hours of light with 3000 lx of light. Survey of surviving, non-surviving and contaminated explants was performed over 5 days over a 30-day period.

Results: Our results have confirmed the difficulties encountered in decontamination for obtaining pure, pathogen-free starting material when dealing with wild species taken from their natural habitats. Significant differences in the duration of use of the sterilizing agent have been identified. In the 2 and 3 minutes exposure of HgCL₂, a number of non-viable explants were reported.

Conclusion: The present study showed a very good sterilization efficiency of *Mentha pulegium* L. at 1 minute in HgCL₂. For a more detailed study of the species, we recommend the use of other sterilizing agents.

Keywords: *in vitro* propagation, wild type, *Mentha pulegium* L., sterilization

P01_07

Bryophytes and larger fungi on the territory of Vrana Park

Galin Gospodinov^{1,2}, Aneta Lambevsk-Hristova¹, Rayna Natcheva¹, Melanya Gyosheva¹

¹Institute of Biodiversity and Ecosystem Research, Bulgarian Academy of Sciences, Department of Plant and Fungal Diversity and Resources, Sofia, Bulgaria

²University of Plovdiv, Department of Plant Physiology and Molecular Biology, Plovdiv, Bulgaria

Aim: The aim of this study is to reveal the diversity of bryophytes, to contribute new data about larger fungi on the territory of Vrana Park, and to evaluate the state of their habitats.

Material and methods: Vrana Park is situated ca. 11 km from the centre of Sofia. The average elevation is ca. 560 m asl.

Field studies were carried out in 2017-2018 using the transect method. Transects were selected in order to cover the entire range of microhabitats.

Results: Bryophyte diversity comprises 59 species of mosses (Bryophyta) and 7 species of liverworts (Marchantiophyta). Four species are of conservation importance. Larger fungi are 28 species (Ascomycota - 3 and Basidiomycota - 25). Lignicolous fungi on dead and living wood (saprotrophs and parasites) prevail. One species is a bryoparasitic fungus.

The habitats in the park comprise wooded areas with plenty of dead wood, single standing trees, grasslands, unpaved roads and paths, rocks and concrete.

Conclusion: The study territory harbours considerable diversity of bryophytes and larger fungi. This is due to the large diversity of habitats. In general, they are in a good state, especially the grasslands, rocky habitats and single standing old trees. Many of the old trees in the forested area however are extremely overgrown by *Hedera helix*, which has a negative impact on the diversity of epiphytic species.

Acknowledgements: This work was held within the project „Investigation of the flora and vegetation: diversity, distribution, biosystematics, dynamics and conservation, Phase II”.

Keywords: ascomycetes, basidiomycetes, bryophytes, lignicolous fungi, Vrana Park

P01_08

Impact of small hydropower plants (sHPP) on the ecological state and macrozoobenthic trophic structure (Iskar River, Bulgaria)

Maria Kerakova, Stefan Kazakov, Emilia Varadinova, Luchezar Pehlivanov

Institute of Biodiversity and Ecosystem Research, Bulgarian Academy of Sciences

Aim: The aim of this study is to assess the impact of small hydropower plants (sHPP) in the middle section of the Iskar River on the ecological status and trophic structure of the macrozoobenthos.

Material and methods: The macrozoobenthic samples were collected during the summer of 2017. An adapted version of the multi-habitat sampling method was used. The sampling was carried out before and after each of the five run-of-the river HPPs. To assess the cumulative effect, macrozoobenthos samples were taken also from two control river sites, located before and after the sHPP cascade. After the samples processing, the zoobenthic taxa were assigned to six functional feeding groups. The ecological classification was done using the standard indices as described in the Ordinance H-4/2012 referred to river type R5.

Results: The ecological state assessment of the river stretches, showed dominance of the moderate and good ecological status. The total number of individuals, found into the river sites after the HPPs, was higher than those in the pool zones. The primary consumer – the scrapers, were the dominant functional feeding group. The qualitative composition and the quantitative parameters of the macrozoobenthic trophic structure from the two control points (before and after the sHPP cascade) demonstrated similar results.

Conclusion: The assessment of the ecological status and the functionality of the macrozoobenthic community, based on the trophic structure, did not show significant cumulative effect from the sHPP exploitation, but the small differences in the surveyed parameters of the community rather reflected the anthropogenic effects on the water ecosystems.

Keywords: macrozoobenthos, functional feeding groups, sHPP

P01_09

Diversity and distribution of benthic diatom assemblages in small and medium-sized Black sea rivers and assessment of their ecological status

Tsvetelina Isheva¹ & Nadya Georgieva Ognyanova-Rumenova²

¹ Institute of Biodiversity and Ecosystem Research, Bulgarian Academy of Sciences, Department of Aquatic Ecosystems, 2, Gagarin str., 1113 Sofia, Bulgaria

² Geological Institute, Bulgarian Academy of Sciences, Acad. G. Bonchev Str., bl. 24, 1113 Sofia, Bulgaria

Aim: The study aimed at investigating the taxonomic composition, ecology and distribution of epilithic diatoms from small and medium-sized Black sea Rivers (river type R11) in Bulgaria. Furthermore, based on the diatom assemblages, the ecological status of these rivers was assessed.

Material and methods: Small (catchment area <100 km²) and medium-sized (100-1000 km²) rivers, which drain into the Black sea are classified according to Bulgarian legislation as national rivers type R11. They are characterized by the extremely high fluctuations of flow, intermittent water regime and strong torrential nature. Diatom sampling, sample treatment, taxa identification and relative abundance, as well as data processing were done following the European standard protocols. The ecological status was established, based on the values of the diatom metric IPS (Specific Pollution Sensitivity index), applying a river type-specific scale for the river type R11.

Results: The epilithic diatom flora was represented by a relative high diversity with some rare and/or poorly known taxa. The rapid pennate diatoms were predominant, followed by the araphid pennate diatoms and last were the group of the centric diatoms. The ecological status of these rivers during the sampling period varied in a wide range.

Conclusion: The results of the study contribute to the knowledge of the diversity, distribution and ecology of the diatom communities from small and medium sized Black sea Rivers and their overall ecological status.

Acknowledgements: The authors wish to thank the “*Programme for support of young scientists and PhD students (2017) in the Bulgarian Academy of Sciences*”, which funded the study.

Keywords: Black sea Rivers, Bulgaria, diatoms, ecological assessment, intermittent rivers

P01_10

What do we know about the vegetation and habitat diversity of Strazhata hill, Northern Bulgaria?

Momchil Nazurov¹, Gana Gecheva¹, Constantin Mardari², Kiril Vassilev³

¹University of Plovdiv “Paisii Hilendarski”, Faculty of Biology, Department of Ecology and Environmental Protection, Todor Samodumov st. 2, 4000 Plovdiv, Bulgaria

²Anastasiie Fătu Botanic Garden, Alexandru Ioan Cuza University, Dumbrava Roşie st. 7–9, 700487 Iaşi, Romania

³Institute of Biodiversity and Ecosystem Research, Bulgarian Academy of Sciences, Department of Plant and Fungal Diversity and Resources, Acad. G. Bonchev st. 23, 1113 Sofia, Bulgaria

Aim: Analysis and summarizing of the existing knowledge about the vegetation and habitat diversity on the territory of Strazhata hill, situated between Sevlievo and Gabrovo.

Material and methods: The study area covers 257.51 km² and only 19% of its territory falls into the NATURA 2000 network in Bulgaria. We checked all phytocoenological publications from Bulgaria and we found only a few related to vegetation and habitat diversity on the territory of the study area. We also used available information about NATURA 2000 sites. Four sites fall into the study area – BG0000263 Skalsko, BG0000214 Dryanovski manastir, BG0000190 Vitata stena and BG0000618 Vidima. In addition, we used the information about the vegetation diversity from the Balkan Vegetation Database (EU-00-019) and the Balkan Dry Grassland Vegetation Database (EU-00-013). Syntaxonomical diversity until now has not been studied following the Braun-Blanquet approach.

Results: A total of 12 habitat types protected by the Habitat Directive 92/43/EEC were found and cover an area of 3650 ha. Woodland vegetation has the widest distribution – 2606 ha (71.4%) as Pannonian-Balkan oak-sessile oak forests (91M0) *Galio-Carpinetum* oak-hornbeam forests (9170), Pannonian woods with *Quercus petraea* and *Carpinus betulus* (91G0) and *Tilio-Acerion* forests of slopes, screens and ravines (9180*). The grassland habitat types cover 1024 ha from the study area such as the Semi-natural dry grasslands and scrubland facies on calcareous substrates (*Festuco-Brometea*) (*important orchid sides) (6210) and the sub-Pannonic steppic grasslands (6240*).

Conclusion: The territory of the Strazhata hill is characterized with different habitat types, which are determined by a wide variety of abiotic conditions (altitude, slope, geology, soil types, etc.). The syntaxonomical and habitat diversity are still poorly studied and future studies are necessary.

Keywords: Directive 92/43/EEC, NATURA 2000 sites, Bulgaria, syntaxa

P01_11

In vitro propagation of *Astracantha thracica* (Fabaceae)

Irena Mincheva, Marina Stanilova, Svetlana Bancheva, Malina Delcheva

Institute of Biodiversity and Ecosystem Research, Bulgarian Academy of Sciences, Acad. G. Bonchev st. 23, 1113 Sofia, Bulgaria

Aim: *In vitro* seed germination and plants production of the Balkan endemic and tertiary relict *Astracantha thracica* (Griseb.) Podl., protected by the Biodiversity Act, included in the Red Data Book of Bulgaria as “Vulnerable”, in “Endangered” habitats.

Material and methods: Seeds were gathered from three populations located in the protected areas Lale Bair, Sliven District, Tarnavski Bakadzhik, Yambol district, and near Vodentzi village, Haskovo district. Seeds, 51 per

population, were treated by 5-time consecutive dips in boiling and ice water to stimulate their germination, and after standard disinfection were germinated on MS medium in a culture room. Seedlings were transferred to pots with soil substrate and *ex vitro* adapted first in a climate chamber, and then in room phytotron.

Results: Seed germination began 5 days after their setting. A part of the seeds were eliminated because of fungal contamination; from the 87 survival, 27 didn't germinate, 6 seedlings were sub-cultured, 13 necrotized, and 41 developed normally and after 8 weeks reached size of 5-8 cm, appropriate for *ex vitro* adaptation. Although the plantlets differed by their growth rate, 80.5% survived in the climate chamber, and 5 of them raised additional shoots. After rapid elongation in the room phytotron 33 plants up to 15 cm high, numbering 18, 10 and 5 respectively from the three populations, are ready to be acclimated in the glasshouse.

Conclusion: Plants will be used for both establishment of *ex situ* collections and reinforcement of *Astracantha thracica* natural populations, according to the Action plan adopted for the species.

Acknowledgments: Authors are grateful to the Bulgarian Enterprise for management of environmental protection activities for the financial support of the research (Contract # 11233/ 10 August 2016).

Keywords: Endemic plants, Relicts, Plant conservation, *Ex situ* collections

P01_12

New records of Bulgarian hypogeous fungi

Boris Assyov¹, Monica Slavova²

¹Institute of Biodiversity and Ecosystem Research, Bulgarian Academy of Sciences, Acad. G. Bonchev st. 23, 1113 Sofia, Bulgaria

²4 Krivolak Str., 1000 Sofia, Bulgaria

Aim: The aim of this work is to contribute to the knowledge on the hypogeous fungi in Bulgaria.

Material and methods: Fungi were collected with the aid of trained dogs and were thoroughly documented. Specimens are preserved in the air-dried state after examination in the living state. The observations of the microscopic features were held with a light microscope. Widely used methods for the study of those fungi were applied.

Results: Eight species were recorded for the first time from the Bulgarian localities, namely *Balsamia polysperma*, *Fischerula macrospora*, *Genea arenaria*, *Jimgerdemannia lactiflua*, *Leucangium carthusianum*, *Pachyphlodes nemoralis*, *Tuber maculatum* and *Tuber magentipunctatum*. The latter is the first finding after the original description of the species and together with *P. nemoralis* they constitute the first records from the Balkan Peninsula. Four genera are also first reported from Bulgaria – *Fischerula*, *Jimgerdemannia*, *Leucangium* and *Pachyphlodes*.

Conclusion: Added to the recent reports, the data presented here show that hypogeous fungi are so far a less studied group in the country, while they apparently harbor significant diversity, which is comparable to that of the other southern European realms.

Acknowledgements: The studies of the first author are held within the project 'Taxonomy, conservation and sustainable use of fungi'. Aurelia Paz Conde and Pascal Chautrand are thanked for their kind advice on part of the collections studied.

Keywords: Balkan mycota, truffle-like fungi, truffles

P01_13

New data on the species of Orchidaceae in the Strandzha Natural Park

Svetla Dalakchieva^{1,2}, Antoaneta Petrova³

¹Regional Historical Museum Burgas, 69 Slavyanska Str., 8000 Burgas

²One Nature Non-profit Organization, 31 "Struma" str., 8001 Burgas

³Botanical Garden, Bulgarian Academy of Sciences, P.O. Box 664 1000 Sofia

Aim: The aim of the study was to collect the data about the distribution and population size of the species of Orchidaceae family in the Strandzha Natural Park. The data were used for a subsequent development of specialized software for the identification of the species and their mapping by professionals and amateurs.

Material and methods: During the period April – October, 28 days of field studies were undertaken. All types of habitats were investigated. Special attention was devoted to the areas with limestone basal rocks. For all the registered localities, geographical and habitat data and data about the population size were collected.

Results: The localities of 24 species were registered. *Dactylorhiza incarnata* is a new species for the Strandzha floristic region. *Himantoglossum jankae*, a species included in Annex IIb of the Directive 92/43 EEC, was documented for the first time for the area of SCI BG 0001007Strandzha, with 5 localities. *Ophrys mammosa* was confirmed for the Bulgarian part of the mountain. New localities of rare and protected species: *Anacamptis laxiflora*, *Ophrys apifera*, *Serapias vomeracea* and *Spiranthes spiralis* were found. All populations of *Anacamptis coriophora* belong to subsp. *fragrans*.

Conclusions: Valuable results for the orchid flora of Stranzha Natural Park were obtained during the field studies. The most numerous species is *Anacamptis papilionacea*, followed by *Serapias vomeracea* and *Anacamptis morio*.

Acknowledgements: The data were collected during the work on the project BG-TR-CBC/025, CB005.1.12.025 "ORCHIS – Orchids researches, conservation and habitats in Strandzha". We thank Mr. K. Popov for the active participation during the field work.

Keywords: *Orchidaceae*, distribution, Strandzha Mts

P01_14

New alien coniferous species in Bulgaria

Antoaneta Petrova¹, Rosen Vassilev², Kamen Bakardzhiev², Boris Assyov³

¹Botanical Garden, Bulgarian Academy of Sciences, P.O. Box 664 1000 Sofia

²Bulgarian Biodiversity Foundation, 39-41 Vestletz Str., 1202 Sofia

³Institute of Biodiversity and Ecosystem Research, Department of Plant and Fungal Diversity and Resources, Bulgarian Academy of Sciences, Acad. G. Bonchev st. 23, 1113 Sofia, Bulgaria

Aim: The introduced ornamental species are the major source for naturalized alien plants. In Bulgaria, where the introduction of exotic trees as ornamentals started at the beginning of the 20th century, naturalization of some species was only recently reported. This encouraged us to take studies on the self-reproduction and naturalization of such species.

Material and methods: Observations on plantings of introduced trees in urban areas were made in different regions in Bulgaria. Data about the population numbers, the age structure of the progeny, the habitat conditions, including their management are collected.

Results: Data about the self-reproduction are reported for 4 species of *Abies* and 2 species of *Cedrus*. The American *A. concolor* was recorded in 4 localities, usually with single individuals; *A. cephalonica*, native to

Greece was recorded in 3 localities, the Caucasian *A. nordmanniana* – in 3 localities and the Western Mediterranean *A. pinsapo* – in 1 locality. The abundant self-seeding of the latter in the Vrana Park since at least 20 year allows considering it naturalized. *Cedrus atlantica* was registered in 5 localities and *C. deodara* – in 1 locality. One of the populations is especially numerous, with more than thousand individuals of different ages, including many seed-reproducing ones.

Conclusions: Six species of Pinaceae emerge as new aliens for Bulgaria. Records mostly come from urban habitats: parks, ornamental and greenery plantings along roadsides and in industrial and villa's estates. In some cases, the progeny is established among the native vegetation.

Acknowledgements: This report is a part of the continuous work on the project “Conspectus of the Bulgarian Vascular Flora”.

Keywords: Alien species, Bulgarian flora, *Abies*, *Cedrus*

P01_15

Parasitoids and inquilines associated with galls of *Andricus quercustozae* (Hymenoptera: Cynipidae) in Bulgaria

Ivaylo Todorov¹, Boyan Zlatkov¹, Anelia Stojanova², George Melika³

¹Institute of Biodiversity and Ecosystem Research, Bulgarian Academy of Sciences, Tzar Osvooboditel 1, Bulevard, Sofia, Bulgaria

²Department of Zoology, University of Plovdiv, 24 Tsar Assen Str., 4000 Plovdiv, Bulgaria

³Plant Health and Molecular Biology Laboratory, Directorate of Plant Protection, Soil Conservation and Agri-environment, National Food Chain Safety Office, Budaörsi u. 141-145, 1118 Budapest, Hungary

Aim: The aim of this study is to review the recently known data on parasitoids and inquilines in galls of *Andricus quercustozae* (Bosc) from Bulgaria and to add new information, obtained from our research in Lozenska Mts. Gall wasp *A. quercustozae* (Bosc) is a West Palearctic species which causes some of the most striking and easy recognizable oak galls in nature. Only agamic females are known, but some authors suppose the existence of unknown sexual generation.

Material and methods: Here, we review the recorded data concerning these gall inhabitants. Furthermore, we collected more than 224 galls from the Lozenska Mts. during the winter-spring periods of 2015-2018. The galls were stored in plastic cups outside to obtain the correct phenological data for all emerging insects.

Results: Amongst the twenty-seventh recently known West Palearctic parasitoids in *A. quercustozae* galls, only 10 species are recorded on this host from Bulgaria. Also, amongst the ninth inquiline species associated to this host, just one – *Synergus umbraculus* (Olivier) (Cynipidae), is found to attack the *A. quercustozae* galls in Bulgaria. Data concerning the parasitoids from the Lozenska Mts. includes 33 specimens belonging to 6 chalcidoid species - *Sycophila biguttata* Swederus and *Eurytoma brunniventris* Ratzeburg (Eurytomidae), *Bootanomyia stigmatizans* (Fabricius), *Torymus auratus* (Müller) and *Torymus cyaneus* Walker (Torymidae) and *Ormyrus nitidulus* (Fabricius) (Ormyridae). All inquiline material belongs only to *S. umbraculus*.

Conclusion: According to a comparison between our results and previously recorded ones, we consider that *Bootanomyia stigmatizans* (Fabricius) (Torymidae) is the most abundant parasitoid species in the *A. quercustozae* galls. However, the abundance of the rest of the species differs significantly between our samples and the data from the literature. One inquiline - *Pammene amygdalana* (Duponchel) (Tortricidae), well known inhabitant in oak galls, is firstly recorded from *A. quercustozae* galls for Bulgaria. A case of co-existence of two inquilines - *P. amygdalana* and *S. umbraculus* is found within a single gall on *Quercus dalechampii* Ten. A new gall association of *Torymus cyaneus* Walker is established. Our results about the inquilines fit well to previously recorded and confirm that only one *Synergus* species - *S. umbraculus*, attacks *A. quercustozae* galls in Bulgaria.

Acknowledgements: We thank the project ANIDIV 3, funded by the Institute of Biodiversity and Ecosystem Research, Bulgarian Academy of Sciences.

Keywords: parasitoids, inquilines, *Andricus quercustozae*, Cynipidae, Bulgaria

P01_16

Ethnobotany and exploitation of medicinal plants in the Rhodope Mountains

Irena Mincheva¹, Michaela Jordanova², Ina Aneva³, Ekaterina Kozuharova¹

¹Faculty of Pharmacy, Medical University of Sofia

²Wild Rodopi, wildrodopi.org, “Akad. Metodii Popov” Str., bl. 48, ent. A, 1113 Sofia, Bulgaria

³Institute of Biodiversity and Ecosystem Research, Bulgarian Academy of Sciences, Acad. G. Bonchev st. 23, 1113 Sofia, Bulgaria

Aim: The aim of this ethnobotanical study is to investigate the traditional home use of medicinal and aromatic plants in the Rhodope Mountains. The emphasis is on *Micromeria dalmatica* Benth., a Balkan endemic.

Material and methods: Semi-structured interviews in 15 localities of the Central and Eastern Rhodopes were performed. People were asked about the plant species used for preparing herbal tea and about the origin of the plants (wild or cultivated). Descriptive statistics was used to determine the relative frequencies of the citations (% of all interviewed informants).

Results: The most used plant species by the people in the Eastern Rhodopes were *Hypericum perforatum* 54%, *Tillia* spp. 46%, *Matricaria chamomilla* 46%, *Thymus* spp. 38% and *Origanum vulgare* ssp. *hirtum* 31%. In the Central Rhodopes the most cited plant species were *Origanum vulgare* ssp. *vulgare* 73%, *Sambucus nigra* 53%, *Thymus* spp. 53%, “White mint” (or “Wild mint, *Micromeria dalmatica*) 53%, *Hypericum perforatum* 46%, *Sideritis scardica* 38%. The major source of plants was recorded to be gathering from the wild. Only few reports of cultivation were documented.

Conclusion: The people in the Central and Eastern Rhodopes are well aware of the medicinal and aromatic plants. There is a trend to use species with conservation status, like *Sideritis scardica*, mostly from cultivation. The Balkan endemic *Micromeria dalmatica* appears to be popular in the Central Rhodopes. We detect a possible hazard for this species. Further investigations are necessary to confirm our findings. A monitoring program and broader cultivation of *Micromeria dalmatica* could be recommended as measures for its sustainable management.

Keywords: Medicinal plants, Rhodope Mts, Herbal tea, *Micromeria dalmatica*

P01_17

New species of *Cristamphidelus* Siddiqi and Vinciguerra, 1991 (Nematoda: Alaimida) from Livingston Island, Maritime Antarctic

Milka Elshishka, Stela Lazarova, Georgi Radoslavov, Vlada K. Peneva

Institute of Biodiversity and Ecosystem Research, Bulgarian Academy of Sciences

The genus *Cristamphidelus* Siddiqi and Vinciguerra, 1991 is represented by eight species with a peculiar distribution, all of them discovered in the southernmost regions of South America (southern Chile and Tierra del Fuego). It is found in the soil and mosses in deciduous forests, pastures, and detritus from roots of ferns.

Characteristic features of the genus are the numerous cuticular longitudinal ridges, the prodelpic female genital system, the S-shaped vagina, sculptured eggshell, long and slender spicules and the presence of gubernaculum in males.

The objective of this work was to study a *Cristamphidelus* species from the Livingston Island, Maritime Antarctic.

Material and methods: Nematodes were extracted from soils and plant materials by a Baerman funnel method, killed by gentle heat and fixed in 4% formalin.

Results: *Cristamphidelus* sp. was associated with different mosses, lichens and soil around *Deschampsia antarctica* Desv. and *Colobanthus quitensis* (Kunth) Bartl. It is most similar with *C. andrassyi* (Vinciguerra and Clausi, 1990) and *C. subantarcticus* (Vinciguerra and Clausi, 1990). From *C. andrassyi* this species differs by position of amphidial aperture, longer female gonad and tail, shape of tail; from *C. subantarcticus* it can be distinguished by the position and shape of the amphidial aperture, longer rectum, more anterior position of vulva, shorter distance posterior end of pharynx - vulva, longer female gonad. DNA data and phylogeny reconstructions showed that the new species is closer to other alaimid species with available sequences in the GenBank.

Conclusion: *Cristamphidelus* sp. is a rare, endemic species, hitherto, it is recorded only from the Livingston Island.

Acknowledgements: The present study was supported by the project №64/27.04.2016, Program for career development of young scientists, Bulgarian Academy of Sciences and ANIDIV 3, M. Elshishka is grateful to the Bulgarian Antarctic Institute for the opportunity to participate in The XXVI Bulgarian Antarctic Expedition and collect new material.

Keywords: Taxonomy, morphology, SEM, 18S, D2-D3 28S rDNA

THEMATIC SESSION II

BIOTIC AND ABIOTIC IMPACT OF THE LIVING NATURE AND MECHANISMS OF ADAPTATION

PL02_01

Invasive alien species – potential cheap resources of plant substances for medicinal use

Ekaterina Kozuharova, Iliana Krasteva, Aleksandar Shkondrov, Iliana Ionkova

Medical University of Sofia, Faculty of Pharmacy, Department of Pharmacognosy

Ailanthus altissima (Mill.) (Simaroubaceae) as well as *Amorpha fruticosa* L. and *Gleditsia triacanthos* L. (Fabaceae) have high tolerance of various habitat conditions and potent propagation ability. These features promote their aggressive invasive behaviour. Additionally, they not only over-compete the local vegetation but suppress the seed development. In the newly invaded habitats they might not have suitable herbivores to control their populations. The only effective enemy might be *Homo sapiens*. Humans are known with their destructive power once an object has become significant for industrial utilization.

The **aim** of this study is to review the research data on: 1) Distribution and invasion level of the mentioned above alien plants in Bulgaria; 2) Ethnobotanical data from their native habitats; 3) Modern investigations of phytochemical constituents and pharmacological activity.

A growing body of scientific literature points to vast therapeutic potential and valuable chemical constituents of these alien invasive plants. The main groups of compounds found in the species are polysaccharides, phenolic acids, coumarins, flavonoids, isoflavonoids, terpenes, sterols, saponins and alkaloids. These species have been

proved to possess various pharmacological activities, incl. cytotoxic and tumour-inhibiting, antioxidant, hepatoprotective, insecticide, analgetic, expectorant, as adjuvants to treat metabolic syndrome, etc. Due to the fact that these are aggressive invasive species, they can provide abundant and cheap resources which can be utilized for medicinal purposes. Exploitation of the biomass for medicinal use might contribute to relieving their destructive impact on the natural habitats and biodiversity.

Keywords: biologically active compounds, pharmacological activity, management

PL02_02

Effect of fungicides on soil microbial community

Galina Radeva¹, Michaella Aleksova^{1,2}, Silvena Boteva², Anelia Kenarova²

¹Roumen Tsanev Institute of Molecular Biology, Bulgarian Academy of Sciences

²Sofia University "St. Kliment Ohridski", Faculty of Biology

Aim: Fungicides play important roles in protecting crop quality and yield in modern agriculture. The present study aims to present the effects of the strobilurin group of fungicides on soil activities and on the structure of soil microbial communities. Our research on the impacts of a model fungicide azoxystrobin (Az) on soil bacterial diversity and the potential of Az to cause antibiotic resistance of soil resistome will be presented as well.

Material and methods: Mesocosm experiment was designed with two different types of agricultural soils (loamy sand and clay loam) treated with increasing Az doses (0.28 mg/kg – 28.93 mg/kg) and incubated for a period of three months. The bacterial community diversity and bacterial biomass were determined using 16S rRNA gene retrieval and real time PCR, respectively.

Results: Our results demonstrated a high bacterial diversity in both soil types on the first day of incubation in untreated and treated soils with the highest dose of Az. Based on the real time PCR outputs, the treatment with Az on the first day in loamy sand and on the 30th day in clay loam soils led to an increased presence of bacteria, as it decreased until the end of incubation.

Conclusion: The impacts of Az on soil bacterial diversity and abundance depended strongly on the applied Az dose, time of incubation and soil properties.

Acknowledgments: This research was supported by the National Science Fund (grant No DH11/6/17).

Keywords: azoxystrobin, soil, 16S rRNA gene, bacterial community diversity

L02_01

Characteristics of the photosynthetic apparatus of *Eryngium maritimum* L. and *Polygonum maritimum* L. - halophytic plants from the dunes of Pomorie Lake (Bulgaria) with different adaptive strategy against soil salinity

Lilia Angelova¹, Albena Ivanova¹, Irina Kosakivska², Lilia Babenko², Svetlana Momchilova³, Sabina Taneva³, Albena Momchilova⁴, Liliana Maslenkova⁴

¹Institute of Plant Physiology and Genetics, Bulgarian Academy of Sciences, Sofia 1113, Bulgaria

²M.G. Kholodny Institute of Botany, National Academy of Sciences of Ukraine, Kyiv 01601, Ukraine

³Institute of Organic Chemistry with Centre of Phytochemistry, Bulgarian Academy of Sciences, Sofia 1113, Bulgaria

⁴Institute of Biophysics and Biomedical Engineering, Bulgarian Academy of Sciences, Sofia 1113, Bulgaria

The **aim** of this study was to investigate the structural-functional peculiarities of chloroplast membranes of halophytic species with different adaptive strategy against soil salinity.

Material and methods: Leaves of the euhalophyte *Eryngium maritimum* L. (Apiaceae) and the glycohalophyte *Polygonum maritimum* L. (Polygonaceae) were collected in August 2016 and 2017 from the sand dunes of Pomorie Lake, (Bulgaria). The pigment content was determined spectrophotometrically. Thermoluminescence (TL) measurements from intact leaves and isolated chloroplast membranes were carried out using computerized equipment. Thylakoid membranes were isolated by standard procedure. The initial oxygen burst was measured using a polarographic oxygen rate electrode. The analysis of fatty acids composition was performed by gas-chromatographic techniques.

Results: The comparative TL glow curves analysis show similar overall TL intensity and oscillation pattern of the main TL B- and AG bands in both investigated plants. The oxygen induction curves (initial oxygen burst) exhibit a biphasic exponential decay thus suggesting some decrease in the proportion of functionally active PSII α centers in thylakoids which could be attributed to the reduced grana formation and dominant operation of stroma situated PSII β centres. The comparative analysis of pigments and fatty acids composition of the thylakoid membranes confirmed the existence of specific quantitative and qualitative differences of euhalophyte and glycohalophyte species.

Conclusion: The results show that the investigated euhalophytic and glycohalophytic species have some specific structural characteristics of their photosynthetic membranes, reflecting the different adaptive strategies to soil salinity but similar capability to maintain an efficient PSII function under environmental conditions in their natural habitat.

Acknowledgements: This work was completed in the frames of the bilateral project between the Ukrainian and the Bulgarian Academies of Sciences (2014-2018).

Keywords: *Eryngium maritimum* L., *Polygonum maritimum* L., fatty acid composition, PSII oxygen-evolving activity, thermoluminescence

L02_02

Comparison of the effects of salt stress on two *Paulownia* lines

Martin Stefanov¹, Ekaterina Yotsova¹, Emilia Gesheva², Velmira Dimitrova^{3,4}, Yuliana Markovska³, Snezhana Doncheva², Emilia Apostolova¹

¹Institute of Biophysics and Biomedical Engineering, Bulgarian Academy of Sciences, Sofia, Bulgaria

²Institute of Plant Physiology and Genetics, Bulgarian Academy of Sciences, Sofia, Bulgaria

³Faculty of Biology, Sofia University "St. Kliment Ohridski", Sofia, Bulgaria

⁴Biotree LTD, 7 Bansko shoes Str., Sofia, Bulgaria

Aim: Comparison of the sensitivity of *Paulownia tomentosa x fortunei* (TF) and *Paulownia elongata x elongata* (EE) to NaCl, depending on the treatment duration.

Material and methods: The plants were grown in Hoagland solutions with different NaCl concentrations. PAM chlorophyll fluorescence, P₇₀₀ photo-oxidation, antioxidant activity (DPPH and FRAP assays, flavonoids) and proline content were used for characterization of the salinity effects on the studied *Paulownia* lines.

Results: The treatment of the plants with high concentration NaCl (≥ 150 mM) for 10 days led to stronger inhibition of the maximum quantum yield of PSII (F_v/F_m), the photochemical quenching (q_p) and the linear electron transport rate (ETR) in TF line in comparison to the EE line. In addition, a restriction of the electron flow from Q_A to plastoquinone was observed. The analysis of the P700 photooxidation revealed that the PSI photochemistry was inhibited after 10 days of salt treatment and high NaCl concentration, but the cyclic electron flow was stimulated, as the effects were stronger in EE line than in the TF line. Data also revealed that the increase of the flavonoids and proline content was more pronounced in the TF than in the EE line.

Conclusion: *Paulownia* TF adapted to concentrations up to 150 mM NaCl for 15 to 25 days, while EE adapted up to 100 mM NaCl for the same duration of the treatment.

Acknowledgements: This work was supported by the Bulgarian Academy of Sciences and the Program for career development of young scientists, BAS (Contract ДФНП-17-135/01.08.2017).

Keywords: *Paulownia*, NaCl, hydroponic system

L02_03

Molecular detection of *Nosema ceranae* and *Nosema apis* in Bulgarian honey bees

Iskra Yankova¹, Daniela Sirakova¹, Boyko Neov¹, Georgi Radoslavov^{1,2}, Peter Hristov^{1,2}

¹Institute of Biodiversity and Ecosystem Research, Bulgarian Academy of Sciences, Department of Animal Diversity and Resources, Sofia, Bulgaria

²Institute of Molecular Biology, Bulgarian Academy of Sciences, Department of Structure and Function of Chromatin, Sofia, Bulgaria

Nosema apis and *Nosema ceranae* are the two main microsporidian parasites causing nosematosis in honey bee *Apis mellifera*.

The **aim** of the present study is to investigate the presence of *Nosema apis* and *Nosema ceranae* in the area of Bulgaria.

Material and methods: The 16S (SSU) rDNA gene region was chosen for analysis. A duplex PCR assay was performed on 108 honey bee samples from three different parts of the country (South, North and West Bulgaria).

Results: The results showed that the samples from the northern part of the country were with the highest prevalence (77.2%) for *Nosema ceranae* while those from the mountainous parts (the Rodopa Mountains, South Bulgaria) were with the lowest rate (13.9%). Infection with *Nosema apis* alone and co-infection *N. apis/N. ceranae* were not detected in any samples. These findings suggest that *Nosema ceranae* is the dominant species in the honey bee in Bulgaria. It is not known when the introduction of *Nosema ceranae* in Bulgaria has occurred, but like in the rest of the world, this species has become the dominant one in Bulgarian *Apis mellifera*.

Conclusion: In conclusion, this is the first report for molecular detection of *Nosema* infection of honey bee in Bulgaria. The results showed that *N. ceranae* is the main *Nosema species* in Bulgaria.

Keywords: Nosemosis, *Apis mellifera*, duplex PCR

L02_04

Biological and statistical comparison of experimental results- good approach for analyzing the adaptive potential of genetically closely related genotypes

Tsveta Angelova¹, Valeria Simeonova², Zhana Mitrovska¹, Stephka Chankova^{1*}

¹Institute of Biodiversity and Ecosystem Research, Bulgarian Academy of Sciences, Sofia, Bulgaria

²Faculty of Mathematics and Informatics, Sofia University "St. Kliment Ohridski", James Bourchier Blvd., 5, 1164 Sofia, Bulgaria

Aim: to compare the biological and statistical data and to identify the most reliable marker/s for the evaluation of the genotype's adaptive potential of three *Phaseolus vulgaris* L genotypes to single and combined treatment with PEG and UV-B irradiation.

Material and methods: Seeds were germinated and grown at standard conditions to the cotyledon phase in a growth chamber. Mild drought stress was simulated by 16% PEG treatment for 24h. UV-B irradiation with 100, 250, 500 J/m² was carried out in BLX 254 UV Cross-linker. Drought and three UV-B doses given on the 12 hour of PEG treatment were applied. Then the plants were grown to the first leaf phase (ten days after the removal of the stress factors) at sterile tap water. After that, measurements of the MDA, Pox, Pro and HSP70B contents were performed.

One-way ANOVA with Tukey multiple comparison tests (GraphPad Prism 6.04 software, San Diego, USA), ANOVA (EXCEL 2007) and Analysis ToolPak were calculated to assess the differences among the samples. To identify the most reliable marker/s the statistical data were summarized with Boolean algebra. Further possible combinations were analyzed using the Boolean operators AND, OR, XOR.

Results: No effect of genotype was shown using the Pox and MDA as markers. Concerning Pro content, an effect of the genotype was found. Dose dependent enhancement of Pro content was measured for the genotype Dobrudjanski 7. Lower Pro accumulation was obtained for Dobrudjanski ran comparing with that in the control. No statistically proven differences with the control were calculated for genotype Dobrudjanski 2 except the sample of 250 J/m² UV-B. The same experimental scheme did not enhance the HSP70B content of the Dobrudjanski 7 genotype (lower/similar to control levels). On the other hand the genotypes Dobrudjanski 2 and Dobrudjanski ran involve higher levels of HSP70B as defense strategy, ten days after a single UV-B and combined treatment.

Data for the four markers were statistically confirmed with Analysis ToolPak (Excel 2007) and ANOVA (Excel 2007). Summarizing the results by Boolean algebra with the Boolean operators AND, OR, XOR, both Pro and HSP70B could be considered as more reliable markers for the adaptive potential.

Conclusion: Our findings show that the combination of biological and statistical comparison of the experimental results is a good approach that can be used to analyze the adaptive potential of genetically closely related genotypes.

Acknowledgements: This study was funded by the projects: DDVU_02/87 “Complex morphometric, physiological, biochemical and molecular assessment of drought tolerance in Bulgarian common bean genotypes (*Phaseolus vulgaris* L.)” and „Ecological and genetic assessment of the environmental - management and strategies for overcoming the risk“ – Bulgarian Academy of Science.

Keywords: *Phaseolus vulgaris* (L.), malondialdehyde (MDA), total peroxides (Pox), proline (Pro), heat shock protein content (HSP70B), Boolean algebra

L02_05

Nurelle D bioactivity depending on the test system

Teodora Todorova¹, Petya Parvanova¹, Abdel-Tawab H. Mossa², Kostadin Kostadinov³, Martin Dimitrov³, Ivan Iliev⁴, Zhana Mitrovska¹, Tsveta Angelova¹ and Stephka Chankova¹

¹Institute of Biodiversity and Ecosystem Research, Bulgarian Academy of Sciences, Sofia, Bulgaria

²Environmental Toxicology Research Group (ETRU), Pesticide Chemistry Department, National Research Centre, Dokki, Giza, Egypt

³Sofia University “St. Kliment Ohridski”, Faculty of Biology, Sofia, Bulgaria.

⁴Institute of Experimental Morphology, Pathology and Anthropology with Museum, Bulgarian Academy of Sciences, Sofia, Bulgaria.

Aim: to evaluate genotoxic, mutagenic and carcinogenic potential of pesticide Nurelle D on different test systems; to compare Nurelle D effects with those of chlorpyrifos as a main constituent of Nurelle D on rats.

Material and methods: Cell cultures of *C. reinhardtii* (WT) and *S. cerevisiae* (strains 551 and D7ts1) in the end of exponential and the beginning of stationary phase were treated with various concentrations of Nurelle D (0.00125 - 0.02% and 0.01-0.6%) for 30 min. Several endpoints were applied: cell survival, “visible” mutations, gene conversion, reverse mutation, mitotic crossing-over as well as Ty1 test for carcinogenicity. Cytotoxic/antiproliferative activity of Nurelle D was evaluated on non-tumorigenic epithelial cell line MCF-10A using standard MTT-dye reduction assay. *In vivo* experiments were conducted to reveal clastogenic potential of chlorpyrifos. Chromosomal aberrations, mitotic index and micronuclei (MN) in polychromatic erythrocytes (PCEs) of rat bone marrow were counted.

Results: Concentrations of Nurelle D that induce 50% lethality were calculated: *C. reinhardtii* - LD₅₀ = 0.0055%, *S. cerevisiae* - LD₅₀ = 0.2% (haploid strain 551) and LD₅₀ = 0.01% (diploid strain D7ts1). No mutagenic effect was found in *C. reinhardtii* in a concentrations range used by us and an increase of gene conversion, reverse mutations and total aberrant in *S. cerevisiae* when concentrations equal or higher than 0.1% were applied.

In vitro experiments show that IC₅₀ varies depending on the incubation time (24h-72 h) in the range of 0.0033% to 0.00265% respectively. Concentrations higher than 0.0008% can reduce cell proliferation of MCF-10A significantly.

In vivo rat studies indicate that treatment with chlorpyrifos can result in a reduction in the mitotic index of about 2-fold, an increase in chromosomal aberrations about 7-fold, and an increase in the total number of micronucleated polychromatic erythrocytes (MMPSE) about 4 fold.

Conclusion: Interspecies and genotype’s variability was obtained comparing concentrations of Nurelle D that determine 50% lethality. Well expressed cytotoxic and genotoxic effects of Nurelle D were revealed even after the treatment with very low concentrations in a test systems *C. reinhardtii* and mammalian cell line MCF-10A. Concentrations equal or higher 0.1% could provoke different genetics events in *S. cerevisiae*. Clastogenic capacity of chlorpyrifos was confirmed *in vivo* experiments with rats. Further experiments should be performed to clarify the mode of action of Nurelle D and both main constituents of this pesticide.

Acknowledgements: This study was funded by the „Strategies for overcoming the genetic risk of anthropogenic pollution with organophosphorus pesticides“- Joint Research Project between BAS and National Research Centre, Arab Republic of Egypt and „Ecological and genetic assessment of the environmental - management and strategies for overcoming the risk“ – Bulgarian Academy of Science

Keywords: *C. reinhardtii*, chlorpyrifos, Nurelle D, *S. cerevisiae*, rats

L02_06

Is there a relationship between DNA repair efficiency, HSPs and genotype resistance?

Daniela Miteva¹, Zhana Mitrovska¹, Nadejda P. Yurina², Stephka Chankova¹

¹Institute of Biodiversity and Ecosystem Research, Bulgarian Academy of Sciences, Sofia, Bulgaria

²Bach Institute of Biochemistry, RAN, Moscow, Russia

Introduction: Different mechanisms of adaptation to environmental stress have been developed by organisms during the evolution: efficient DNA repair systems, cell membrane stability, induction of heat shock proteins (HSPs), particularly chaperons etc.

The aim of this study is to analyze the contribution of DSBs DNA repair efficiency and HSP70B to the formation of genotype resistance.

Material and methods: Unicellular green algae are used: *Chlamydomonas reinhardtii* - 137C WT, UVS10 *rec* and UVS14 *mismatch* repair deficient and *Chlorella* - *Chlorella vulgaris* (Antarctic), *Chlorella vulgaris* strain

8/1(Thermophilic) and *Chlorella kessleri* (Mesophilic). The algae are cultivated on liquid TAP medium under standard conditions in the growth camera Phytotron GC 40. The cell suspensions at the end of the exponential/beginning of the stationary phase are used.

Chlorella species are irradiated with UV-B ($\lambda = 312$ nm) in BLX-254, Life Technology, UV crosslinker. The irradiation is in a dose range of 50, 100, 250, 500, 1000 J/m². The genotype resistance is determined on the basis of a spot-test, microcolonies assay, photo-reactivation sectors and growth rate.

The *Chlamydomonas* strains are treated with zeocin that can cause different damages including double strand breaks in DNA, oxidative stress and cell death.

The DSBs induction and repair are efficiency measured by CFGE. The HSP70B are determined using gel-electrophoresis and Western blotting

Results: Our results show that doses higher than 250 J/m² are bioactive for *Chlorella* species. Species photo-resistance to UV-B evaluated on the basis of LD₂₀, LD₅₀ and LD₉₀ and sector of photo-reactivation is different: *Chlorella vulgaris* > *Chlorella vulgaris* 8/1 > *Chlorella kessleri*. Species differ in their capacity to overcome the harmful effect of the UV-B measured as DSBs induction and repair efficiency. Strains UVS-10 *rec* and UVS-14 *mismatch* repair deficient have significantly lower capacity to repair DSBs induced by zeocin comparing with strains 137C. The increased levels of HSP70B content 2 h after zeocin treatment are measured for strains UVS-10 and UVS-14. Strains with WT resistance - 137C + WT respond to zeocin induced oxidative stress later - on the 4th hour after the treatment.

Conclusion: Based on the results obtained it could be said that HSP70B and both repair systems - *rec* and *mismatch* contributed to the formation of genotype resistance of investigated by us strains.

Acknowledgements: This study was funded by the project: “Antarctic algae – a model-system for resistance to oxidative stress”. D-002-317 and the cooperation between BAS and RAS.

Keywords: unicellular green algae, DNA repair efficiency, HSPs, genotype resistance

L02_07

Genotoxic and mutagenic effects of *Narcissus triandrus* L. total extract on *Chlamydomonas reinhardtii*

Petya Parvanova, Zhana Mitrovska, Boriana Sidjimova, Milena Nikolova, Strahil Berkov and Stephka Chankova

Institute of Biodiversity and Ecosystem Research, Bulgarian Academy of Science

Aim: to evaluate the genotoxic and mutagenic potential of total leaves extract of *Narcissus triandrus* L. on a test-system *Chlamydomonas reinhardtii*.

Material and methods: *N. triandrus* extract was derived and prepared by prof. Berkov's team.

C. reinhardtii strain 137 C+ (WT) was used as a test system. Cultures at the end of exponential and the beginning of stationary phases were treated for 2 hour with several concentrations - 250 µg/ml, 500 µg/ml, 750 µg/ml and 1000 µg/ml. The survival fraction was analysed on the basis of colony forming ability. The test of the visible mutations was performed to reveal the mutagenic capacity of the extract.

The results of the treated samples were compared with those of the control ones - Sager-Granick liquid medium (SG), DMSO (0.1%) and PQ (5 µM) as a positive control.

Three levels of lethality were calculated - LD₂₀; LD₃₇ and LD₅₀.

Results: No statistically significant genotoxic effect was found using 250 µg/ml *N. triandrus* L total extract. The treatment with concentrations of 500 µg/ml, 750 µg/ml and 1000 µg/ml resulted in decreased colony forming ability in a statistically significant way compared with both negative controls – SG and DMSO. The genotoxic capacity of *Narcissus triandrus* L. leaves extract was similar to that of 5 µM PQ. Concentrations 750 µg/ml and

1000 µg/ml were obtained to have well expressed genotoxic capacity. Using the Reed and Muench method three levels of lethality $LD_{20} = 352$ µg/ml; $LD_{37} = 482$ µg/ml and $LD_{50} = 598$ µg/ml were calculated. No mutagenic effect was revealed.

Conclusion: Statistically significant genotoxic effect was found after the treatment with concentrations of *N. triandrus* leaves extract higher than 250 µg/ml for 2 hr without any mutagenic capacity.

Acknowledgements: This study was funded by the „Program for career development of young scientists, Bulgarian Academy of Science“ and „Ecological and genetic assessment of the environmental - management and strategies for overcoming the risk“ – Bulgarian Academy of Science.

Keywords: *Chlamydomonas reinhardtii*, extract, genotoxicity, mutagenic, *Narcissus triandrus* L.

P02_01

European roe deer (*Capreolus capreolus*) as a biomonitor for the current environment heavy metals contamination in an agricultural region in Bulgaria

Georgi Markov*, Atidzhe Ahmed

Institute of Biodiversity and Ecosystem Research, Bulgarian Academy of Sciences

Aim: The aim of the study was to evaluate the current contamination of liver and kidney tissues of the European roe deer (*Capreolus capreolus*) inhabiting a typical agricultural region in Bulgaria by priority pollutants of the heavy metal group (Pb, Cd, Cu and Zn).

Material and methods: All investigated 15 adult male roe deers (between 2 and 5 years) inhabited an area covered by forests and arable lands. The roe deers' liver and kidneys were used as test systems for determination of the analytic concentrations of the studied heavy metals by an inductively coupled plasma atomic emission spectrometry (ICP-AES) using a Perkin Elmer Optima 7000 DV.

Results: The concentrations [mg/kg dry tissues] of the tested metals, specific for each organ, characterized by the width of the range about the median that includes 95% of the cases demonstrate the limits of their variability. In the liver, the limits of variability for Cu were from 25.99 to 269.85; for Zn from 70.81 to 1115.63; for Pb from 0.81 to 9.76 and for Cd from 0.86 to 5.31. In the kidneys respectively they were for Cu from 46.49 to 83.07; for Zn from 116.29 to 1542.52; for Pb from 0.71 to 8.40 and for Cd from 8.11 to 74.92.

Conclusion: These results create a baseline for estimation of the current heavy metal accumulation in roe deer and provide an opportunity to use it as a bioindicator of the future potential anthropogenic negative influence on the environment in the agricultural regions of the country, under the conditions of modern agricultural activities in them.

Acknowledgements: This study was supported by the project № ДФНП-17-50/26.07.2017 “Study of the roe deer (*Capreolus capreolus* L.) in Bulgaria as accumulation bioindicator of heavy metals in the environment” developed in the “Program to support young scientists and doctoral students of BAS-2017”.

Keywords: weight of tissue samples, biomonitoring, European roe deer, *Capreolus capreolus*

P02_02

Interrelationship between fresh and dry weight of internal target organs of roe deer (*Capreolus capreolus*): application for the purposes of biomonitoring of the environment

Georgi Markov*, Atidzhe Ahmed

Institute of Biodiversity and Ecosystem Research by Bulgarian Academy of Sciences

Aim: The present research aims to evaluate the nature and the degree of the relationship between fresh and dry weight of the internal organs (liver and kidneys) of the European roe deer (*Capreolus capreolus*), which would allow to comparatively assess the analytical concentrations of xenobiotics in them, obtained by different methods of preparation of these tissues for analysis.

Material and methods: The tissues from the liver (n=38) and kidneys (n=33) were taken from adult roe deers (over 2 years) inhabiting areas with different anthropogenic impact in Bulgaria. All calculations in this study were performed using the statistical package STATISTICA (StatSoft Inc. 2011).

Results: Under laboratory conditions, the fresh and the dry weight of the investigated tissue samples were weighed with accuracy to 0.001g. The established correlation ($R = 0.981$ in the liver and $R = 0.884$ in the kidney) and regression relationships (linear regression models) between the fresh and the dry weight of the studied samples, as well as the percentage of liquid content in them (71.75% in the liver and 79.75% in the kidney), allow a comparison of the analytical concentrations of heavy metals measured in fresh and in dry tissues of the liver and kidney of the roe deer.

Conclusion: This new opportunity for the comparative assessment of the analytical concentrations of xenobiotics in examining target organs strongly supports the comparative analysis of the accumulation of priority natural pollutants in the roe deer's body and its use as a bio-indicator for the assessment of the environmental quality in both geographical and time aspects.

Acknowledgements: This study was supported by the project № ДФНП-17-50/26.07.2017 “Study of the roe deer (*Capreolus capreolus* L.) in Bulgaria as accumulation bioindicator of heavy metals in the environment” developed in the “Program to support young scientists and doctoral students of BAS-2017”.

Keywords: weight of tissue samples, biomonitoring, European roe deer, *Capreolus capreolus*

P02_03

Growth light intensity effect on the structural stability of LHCII in pea thylakoids

Nia Petrova, Svetozar Stoichev, Svetla Todinova, Stefka Taneva, Sashka Krumova

Institute of Biophysics and Biomedical Engineering, Bulgarian Academy of Sciences

Aim: The thylakoid membrane of higher plants is a very flexible system dynamically responding to the changes in the environmental conditions with structural rearrangements of its constituents and protein conformational changes. A main player in those processes is the major light harvesting complex of photosystem II (LHCII) that is also the most abundant thylakoid protein. LHCII has both structural and functional role - it maintains the membrane stacking and optimizes the light-harvesting. The aim of the study is to examine how the structural

arrangement of LHCII in thylakoids isolated from pea plants grown at normal and low light intensity affects its conformational stability.

Material and methods: *Pisum sativum* cv. Ran I plants were grown hydroponically at 150 (normal) and 20 (low) $\mu\text{mol photons/m}^2\text{s}$ light intensity for 14 days. Thylakoid membranes were consequently isolated and characterized by circular dichroism and differential scanning calorimetry.

Results: Low light growth induced higher chlorophyll *b* content, more ordered arrangement of the LHCII complexes and tighter membrane stacking. Those features were associated with downshift of the thermal stability of the complex as compared to normal light grown controls.

Conclusion: Our data demonstrate that the structural organization of the thylakoid membranes strongly affects the LHCII structural stability. Thylakoids prepared from low light grown plants are more ordered compared to the control in both vertical and horizontal direction which results in alteration of the conformation of LHCII and reduction of its thermal stability.

Acknowledgements: This work is supported by project DFNP 17-138 (granted to Nia Petrova), Program for career development of young scientists, Bulgarian Academy of Sciences.

Keywords: thylakoid membrane, differential scanning calorimetry, circular dichroism, major light-harvesting complex of photosystem II

P02_04

Community structure and individual reaction of Chironomidae (Diptera) larvae in anthropogenically impacted sites of the Danube River (NE Bulgaria)

Mila Ihtimanska, Julia Ilkova, Paraskeva Michailova

Institute of Biodiversity and Ecosystem Research, Bulgarian Academy of Sciences

Aim: The aim of the study is to establish the response of Chironomidae at individual and community level to the anthropogenic pressure in stations of the Danube River.

Material and methods: The sampled sites were located after tributary Rusenski Lom (1) and in the industrial area of the cities of Ruse (2) and Silistra (3). A total of 14 genera of the family Chironomidae were determined by external morphology and 6 species – cytotaxonomically. Initially physicochemical parameters, nutrients and heavy metals in the water and sediment were used for the analyses. The morphological deformities were analyzed in percentage. Spearman's *r_s* coefficient was used for a correlation between the taxa, percentage of deformities and environmental variables.

Results: The low values of the Shannon-Wiener's diversity index ($0 \div 2$) showed a disturbed chironomid community. At sites 1 and 2 the concentrations of some heavy metals and nutrients in water and sediments exceeded the reference data. At both sites the genera *Cricotopus* sp., *Paratanyratus* sp. and *Polypedilum* sp. occurred in high percentage. The species: *Cricotopus* sp. and *Polypedilum* sp. showed correlations ($P < 0.05$) with alkalinity, Cu, nitrates in water and sediments. The percentage of the deformities correlated with the phosphates in the sediment ($P < 0.05$).

Conclusion: The results showed the anthropogenic impact on the Chironomid taxa. The potential of midge deformities as a biomonitoring tool is shown, however they should be used very carefully because a number of abiotic and population factors can affect their occurrence.

Acknowledgements: The authors thank to "Program for supporting young scientists and PhD students – 2017" of Bulgarian Academy of Sciences.

Keywords: Chironomidae, Danube, deformities, environmental variables, anthropogenic impact

P02_05

Morphogenesis and developmental patterns affect polyphenolics production and endogenous stress hormones in *Artemisia alba* Tura *in vitro*

Kalina Danova¹, Vaclav Motyka², Petre Dobrev²

¹Institute of Organic Chemistry with Centre of Phytochemistry, Bulgarian Academy of Sciences, Sofia, Bulgaria

²Institute of Experimental Botany, Czech Academy of Sciences, Prague, Czech Republic

The present work **aims** at the elucidation of the effect of the morphological development of *Artemisia alba* Tura on the production of polyphenolics and the content of endogenous stress hormones *in vitro*.

Material and methods: Plant growth regulators treatments were applied as follows: GAIP_0 – PGR-free control; GAIP_1 - 0.5 mg/l IBA; GAIP_2 - 1.0 mg/l IBA; GAIP_3 - 0.2 mg/l BA + 0.5 mg/L IBA and GAIP_4 - 0.2 mg/L BA + 1.0 mg/L IBA. Total phenolic and flavonoid contents were assayed colorimetrically. The endogenous levels of abscisic acid (ABA) and its catabolites (dihydrophaseic acid, phaseic acid, abscisic acid-glucose ester, neophaseic acid and 9-hydroxy-abscisic acid), jasmonic acid (JA) and jasmonic acid-isoleucine, as well as (SA) salicylic acid were analyzed by LC/MS.

Results: irrespectively of the different concentrations, PGR treatments of *A. alba in vitro* resulted in the development of two main morphotypes. While PGR-free control and IBA treated plants were characterized with the development of both aerial and root tissue, the combined IBA and BA treatments resulted in root inhibition and callusogenesis at the explant base. The root inhibited morphotype displayed elevated polyphenolic levels and a drop of SA and ABA and its pool. On the contrary, the content of JA and its pool were elevated in the latter group of plants. Noteworthy, the observed dependencies were related to the developed morphological type of the plants but not to the concentration of the plant growth regulators applied.

Conclusion: Secondary metabolites production was shown to be affected by the developmental patterns and morphogenesis of *A. alba in vitro*. Such dependencies could be used as a tool for the biotechnological secondary metabolite production of medicinal and aromatic plants without performing genetic transformations.

Acknowledgements: This work is supported by the bilateral agreement program between the Bulgarian and Czech Academies of Sciences

Keywords: *Artemisia alba* Tura, *in vitro* morphotypes, endogenous abscisic acid, salicylic acid, jasmonates

P02_06

Changes of antioxidative enzymes during natural regeneration in deciduous forests

Vasil Chavgov¹, Detelina Petrova¹, Miroslava Zhiponova¹, Ganka Chaneva¹, Adriana Marinova², Svetoslav Anev², Nikolina Tzvetkova²

¹ Sofia University “St. Kliment Ohridski”, Faculty of Biology

² University of Forestry

Altitude and location are among the factors that strongly influence the physiology of deciduous trees. As we have previously shown, the common beech and hornbeam underwent some changes in the antioxidant status, non-enzymatic protective mechanisms and pigment content of the leaves, depending on their exposure.

Aim: Two widespread species - *Fagus sylvatica* L. and *Carpinus betulus* L., were investigated in order to determine the impact of these ecological factors during the natural regeneration in deciduous forests, with a special emphasize on the changes in the antioxidative enzyme defense system.

Material and methods: The observations were carried out at the Petrohan Experimental Forest Range, at three different altitudes (680, 1050 and 1450 m asl). The activity of superoxide dismutase (SOD) was measured according to Beauchamp & Fridovich (1971), the activity of catalase (CAT) – according to Aebi (1984). Peroxidase (PER) activity was determined by the method described by Hart et al., 1971.

Results: Common beech and hornbeam reacted differently to the changes in the exposure and altitude. Antioxidative enzymes showed increased activities in the beech leaves at the full sun exposure, particularly pronounced at 680 and 1400 m asl. The highest levels of SOD and CAT activities were measured at 1450 m asl, while PER activity was higher at 680 m asl. In the hornbeam saplings grown at the forest's patches, the full sun exposure caused an opposite trend – SOD and PER decreased their activities. Meanwhile, CAT activity remained only slightly enhanced.

Conclusion: The full sun exposure could probably trigger the activation of the enzyme protection system as a response to the oxidative stress conditions. That assumption was confirmed by the strongly enhanced antioxidative activities in the common beech. However, the opposite reaction of the hornbeam enzyme system suggested that different deciduous species would have developed a variety of physiological mechanisms against identical stress factors.

Acknowledgements: The study was supported by grant № 159/2017 of Sofia University of Forestry.

Keywords: *Carpinus betulus*, *Fagus sylvatica*, superoxide dismutase, catalase, peroxidase

P02_07

Dynamics of the health status of coppice oak forests in Southwestern Bulgaria

Nikolay Zafirov, Krasen Banov

University of Forestry, Sofia, Bulgaria

Aim: Climate anomalies, air pollution and biotic stress factors have caused several periods of decline (dieback) of different forest types in Bulgaria and in Europe as a whole during the last decades. In order to obtain up-to-date information on the development of this process, the aim of this study is to analyze the dynamics of the health status of coppice oak forests in Southwestern Bulgaria and to identify the main stress factors in them.

Material and methods: The research is carried out in oak forest stands located in 4 State Forestries in the Bulgarian Southwestern State Enterprise. Dendrochronological analysis is predominantly used, which is combined with defoliation assessment, macroscopic and microscopic phytopathological analyzes.

Results: Representative radial increment chronologies for the main oak species in the studied area are developed, which reflect the dynamics of their health status. The obtained coefficients of determination by multifactor regression analysis for the influence of temperature and precipitation regimes on it are high ($R^2 > 50\%$). The results are related also to the management history of the forest stands.

Conclusion: The health status of the studied coppice oak forests was the most deteriorated in the mid-20th century and after the early 1980s. In some of them the last stress period continues until the end of the analyzed period. The most unfavorable climatic conditions for them are associated with low precipitation combined with high air temperatures. They coincide with the determined stress periods, which show that unfavorable temperature-precipitation regime is the main predisposing stress factor in these stands.

Acknowledgements: This study was supported by Contract No. 29/23.03.2017, topic under the Scientific Research Sector in the University of Forestry with code 914: “Justification and implementation of regional forestry systems for management of the coppice oak forests in the territorial scope of the South-Western State Enterprise”.

Keywords: dendrochronology, oak decline, coppice forests, *Quercus petraea* (Matt.) Liebl., *Quercus frainetto* Ten., *Quercus cerris* L.

P02_08

Impacts of the fungicide azoxystrobin on soil bacterial communities

Michaela Aleksova^{1,2}, Jordan Manasiev⁴, Silvena Boteva², Mladen Popov^{1,3}, Anelia Kenarova², Galina Radeva¹

¹“Roumen Tsanev” Institute of Molecular Biology, Bulgarian Academy of Sciences

²Sofia University “St. Kliment Ohridski”, Faculty of Biology

³University of Chemical Technology and Metallurgy

⁴Biovet Botevgrad, 2140 Botevgrad

Aim: Azoxystrobin is a strobilurin fungicide, which is one of the most popular chemicals, used for control of fungal plant pathogens. Azoxystrobin is effective against Ascomycota, Deuteromycota, Basidiomycota and Oomycota, inhibiting their mitochondrial respiration and spore germination. The competition between bacteria and fungi for nutrient resources or the usage of destroyed fungal biomass as a substrate by bacteria may indirectly impact soil bacterial communities. The aim of the study is to evaluate the indirect effect of the fungicide azoxystrobin on soil bacterial communities.

Material and methods: Azoxystrobin was added in increasing concentrations (0.28 mg/kg; 14.46 mg/kg; 28.93 mg/kg) to agricultural soils (loamy sand and loamy clay) in a mesocosm experiment. For both soil types an untreated soil was used as a control. To determine the effect of azoxystrobin on bacterial community abundance 16S (bacteria) rRNA gene copy number qPCR analysis was performed. The fungicide effect was studied for a period of three months.

Results: In loamy sand a stimulating effect on the bacterial amount after treatment with azoxystrobin on the first day was observed. To the 30th day their amount decreased considerably and remained nearly constant to the 90th day. In loamy clay the stimulating effect was observed on the 30th day after which the bacterial amount decreased.

Conclusion: Our results show that the bacterial abundance was affected by the dose of the applied fungicide, the incubation time and the soil type.

Acknowledgments: This research was supported by the National Science Fund (grant No DH11/6/17).

Keywords: azoxystrobin, 16S rRNA gene, qPCR

P02_09

Protein/RNA ratio of quiescent and exponentially grown cells of *Saccharomyces cerevisiae*

Anna Tomova, Anna Kujumdzieva, Ventsislava Petrova

Sofia University “St. Kliment Ohridski”, Biological Faculty, Laboratory “Applied Microbiology”

Aim: The protein/RNA ratio is directly correlated with the growth rate and could be independent of the developmental state of the microorganisms. The importance of this ratio follows from the mechanism of growth as the formation of new biomass corresponds to the protein synthesis (measured by the RNA content).

Material and methods: Two *S. cerevisiae* strains, having different specific growth rates and ploidy were used – a diploid NBIMCC 584 and a haploid BY4741 one. They were grown for 168 h in YPD medium and samples were withdrawn at the exponential and late stationary phase. Quiescent cells were separated by percoll density-gradient centrifugation. To fully define the cell states and kinetic parameters, cellular protein and RNA content were accessed as central targets defining a cell’s phenotype.

Results: An approach is presented to simultaneously and specifically measure the protein and RNA in lysates from cell cultures of logarithmic and G_0 population as a marker for cell functions and responses. The physiological state of the yeast culture is determined by a set of kinetic parameters. The comparison of the RNA protein ratio in actively proliferating and quiescent *S. cerevisiae* cells showed that the G_0 cells have a significantly lower coefficient (0.625) than the proliferating ones – 12.5.

Conclusion: The most important variable for single cell organisms during the course of adaptation is the growth rate (GR) of a population. In this respect it was assumed that the joint interpretation of key kinetic parameters (μ^{\max} , $Y_{X/C}$, $Y_{G0/C}$, q_s , q_{G0}) and cellular protein/RNA ratio could be successfully used for the fitness assessment of a given yeast genotype over time.

Acknowledgments: This work was supported by a grant from the National Science Fund, Ministry of Education and Science, Project № ДН11/10.

Keywords: *S. cerevisiae*, quiescence, kinetics

EP02_01

High-light-inducible small stress proteins and their localization in the chlorophyll-protein complexes of cyanobacteria

Lubov Sharapova, Yuliya Oleskina, Nadezhda Yurina

Bach Institute of Biochemistry, Research Center of Biotechnology of the Russian Academy of Sciences, 33, bld. 2 Leninsky Ave., Moscow 119071, Russia

Aim: The association of high-light-inducible (HliA/HliB) small stress proteins with photosystem 1 (PS1) complexes of the cyanobacteria was studied to understand their function.

Material and methods: Two cyanobacteria *Synechocystis* sp. PCC 6803 and *Arthrospira platensis* were studied. Chlorophyll-protein complexes were fractionated by two-dimension polyacrylamide gel electrophoresis (2D PAGE). Proteins identification was performed by MALDI-TOF mass spectrometry and by Western-blot analysis.

Results: The trimers and monomers of photosystem 1 (PS1), dimers and monomers of the photosystem 2 (PS2), the cytochrome complex, ATP-ase, the NAD (P) H-quinone-oxidoreductase and zone of the free protein were identified by 2D PAGE. The HliA/HliB proteins were associated with the chlorophyll-protein complexes and were shown in the free protein zone by immunoblotting. Hli proteins associated with trimers PS1, complexes of PS2 and monomers PS1. Hli proteins were also found in the free proteins zone.

Conclusion: The association of HliA/HliB proteins not only with PSI trimers, but also with PS1 monomers and PS2 complexes was shown. This fact suggests a universal role of these proteins in the protection of the photosynthetic apparatus from light stress.

Acknowledgements: This work was supported by the Molecular and Cell Biology and post-genomic technologies Program №18 of the Presidium RAS and by the RFBR (project No.16-04-01626A).

Keywords: high light-inducible proteins, light stress, photosystem 1, cyanobacteria

THEMATIC SESSION III

ECOSYSTEM RESEARCH, SERVICES AND ECOLOGICAL AGRICULTURE

PL03_01

Mapping and assessment of the condition and ecosystem services of inland wetlands in Bulgaria outside the Natura 2000 ecological network

Nevena Ivanova

Institute of Biodiversity and Ecosystem Research, Bulgarian Academy of Science

The project "**Wetlands Ecosystem Services Mapping and Assessment in Bulgaria**" (WEMA) was funded under the Program BG03 Biodiversity and Ecosystems under, Call BG03.02: Mapping and Assessment of Ecosystem Services of Financial Mechanism of the European Economic Area 2009-2014. The project organized and analyzed ecological and biological scientific information on wetland ecosystems outside the Natura 2000 network in Bulgaria. The collected information will be integrated into the Information System of the National Biodiversity Monitoring System of the Executive Environment Agency. The project was implemented according to the "Methodology for assessment and mapping of Wetland ecosystems condition and their services in Bulgaria", part B7 which is developed under the project "Methodological Support for Ecosystem Services Mapping and Biophysical Valuation (MetEcoSMap)". Object of mapping and assessment of the condition and services provided were the following subtypes of "inland wetlands" ecosystems: 1 / Valley mires, poor fens and transition mires (D2); 2 / Base-rich fens and calcareous spring mires (D4); 3 / Sedge and reedbeds, normally without free-standing water (D5). The main results are:

- 285 "Inland Wetlands" ecosystems identified and mapped through GIS and field studies;
- The condition of identified ecosystems is assessed by direct measurement or analysis of available data on plant and animal diversity, soils, waters, fires, dumping-grounds and invasive species;
- Assessed provisioning, regulating/maintenance and cultural ecosystem services;
- Individual maps of sub-types of ecosystems, their condition and the ecosystem services they provided was made;
- The information gathered and the assessments made are entered in a database;
- Under the project has published four press releases on the BTA website, two brochures for the project activities (one of which are in English), a book for the project presenting the project results, two project seminars were accomplished, and two films are made - one for the general public and the other is intended for the more specialized audience (scientists, lecturers, experts in the EEA, RIEW).

The results of the project will benefit the Ministry of Environment and Waters, the Environmental Executive Agency, the RIEW, the Basin Directorates, regional and local authorities, water and sewerage companies, non-governmental organizations, business, researchers, students and wetland users. For more information: <http://www.iber.bas.bg/sites/default/files/projects/WEMA> and nevena759344@gmail.com.

Keywords: inland wetlands, mapping, condition, ecosystem services

PL03_02

Mapping of ecosystems types, conditions and their services. Challenges and recent achievements in the Bulgarian Black Sea region

Ventsislav Karamfilov, Dimitar Berov, Valko Biserkov, Georgi Daskalov, Stephania Klayn

Institute of Biodiversity and Ecosystem Research, Bulgarian Academy of Sciences

Action 5 of the EU Biodiversity Strategy to 2020 calls Member States to map and assess the state of ecosystems and their services in their national territory with the assistance of the European Commission of Bulgaria based on existing data was carried out.

Within the framework of the Program BG03-02 "Mapping and Evaluation of Ecosystem Services" of the Financial Mechanism of the European Economic Area (FM/EEA) in Bulgaria started the development of the National Methodological Framework for Mapping and Evaluation of Ecosystems and their services on the 9 main types of ecosystems in Bulgaria, including for marine ecosystems. Finally, the Methodology was applied and under contract № Д-33-87/23.08.2016 an initial mapping of marine ecosystems types, conditions and their services in the EEZ (200 nautical miles from the coast line) was carried out.

The results support the findings that natural biodiversity is a basic factor that supports and maintains the marine ecosystem functions, respectively services especially those that might be crucial to human well-being (e.g. provisioning services such as food supply, as well as regulating and supporting services such as nutrient cycling, regulation of floods and disease control. Therefore, conservation of natural biodiversity in naturally preserved habitats looks to be the only way for a successful sustainable management of ecosystem services. Thus, ecosystem services should not be considered only as the benefits people obtain from ecosystems but also as structures that provide facilities for maintenance of natural biodiversity.

L03_01

Macrophytes-based ecological status assessment of the Dragomansko and Aldomirovsko marshes

Teodora Ilieva¹, Gana Gecheva¹, Kiril Vassilev²

¹University of Plovdiv "Paisii Hilendarski", Faculty of Biology, Department of Ecology and environmental protection, Plovdiv, Bulgaria

²Institute of Biodiversity and Ecosystem Research, Bulgarian Academy of Sciences, Department of Plant and Fungal Diversity and Resources

Aim: Analysis of the aquatic macrophytes composition and assessment of the ecological status of Dragomansko and Aldomirovsko marshes

Material and methods: Aquatic macrophytes survey was carried out in 2017 between July and September within belt transects. In each site usually a 100 m long section was studied following a zigzag pattern. The abundances of all single species were registered using a semi-quantitative five-level scale: 1 = very rare, 2 = infrequent, 3 = common, 4 = frequent, 5 = abundant, predominant, similar to the commonly used DAFOR scale (Dominant, Abundant, Frequent, Occasional, Rare) for bryophyte communities. Field records also included estimation of abiotic parameters: substrate type, maximum depth of macrophytes colonization, etc.

Results: The macrophyte species composition in both marshes was high in the studied period. Thirty-three species were recorded in the Aldomirovsko marsh, among them more than 50% helophytes. Fifty species were registered for the Dragomansko marsh, of which helophytes represented about 30%. The most common taxa for both water bodies were *Carex riparia*, *Typha latifolia* and *Scirpus lacustris*.

Floating species include *Ceratophyllum demersum*, *C. submersum*, *Hydrocharis morsus-ranae*, *Lemna minor*, *L. trisulca*, *Myriophyllum verticillatum* and *Potamogeton lucens*.

The ecological status assessment revealed that the Dragomansko marsh was in good status, while the Aldomirovsko one was assessed as moderate.

Conclusion: Macrophyte flora is a key parameter on which the lake assessment is based, and is a reliable indicator for the changes of the ecosystem's dynamics. Both marshes supported rich macrophyte communities. Considering the lack of previous studies in the selected water bodies, this study presents a basis for specific conditions within the lake type for future researches regarding the ecosystem potential, services and sustainable development of protected wetlands in Bulgaria.

Keywords: macrophytes, marshes, wetland

P03_01

Identification of Ecosystem Types in High Mountain Areas: A case study in the Rila Mountain

Kostadin Katrandzhiev¹, Svetla Bratanova- Doncheva¹, Stoyan Nedkov²

¹Institute of Biodiversity and Ecosystem Research, Bulgarian Academy of Sciences

²National Institute of Geophysics, Geodesy and Geography

Aim: High mountain ecosystems are the most sensitive to climate change at both regional and global scales. Their study could help society to develop mechanisms for adaptive management and policy making. It is necessary to gain enough knowledge about their condition in order to keep their integrity, resilience and ability of self-maintenance. The aim of this study is to present the first step of identification and mapping of different ecosystem types and subtypes in high mountain areas – Rila Mountains.

Material and methods: By a set of criteria a high mountain area in Bulgarian Rila Mountains was chosen for our research. It covers the South West parts of Rila Mountains with the highest point the Ezernik peak (2485 m). Using remote sensing – orthophoto and UAV (Unmanned Aerial Vehicle) images along with computer software (ArcMap version 10.1), literature and fieldwork we identified and mapped the different ecosystem types presented in the chosen study area.

Results: Our efforts resulted in the identification of three main ecosystem types and more than 90 ecosystem subtypes. All of them were classified using the MAES classification and Methodology for assessment and mapping of woodland and forests ecosystems condition and their services in Bulgaria (2017). The results are presented as maps of high mountain ecosystems in the case study area.

Conclusion: The high mountain ecosystems condition assessment could help us to manage the threats arising from the climate change. Making the first step of the ecosystem's type identification and mapping in SW Rila Mountains successfully, we can proceed to their condition assessment.

Acknowledgements: This study was supported by project: Methodological support for ecosystem services mapping and biophysical valuation (MetEcosMap), funded by FM of EEA (2015-2017).

Keywords: High mountain ecosystems, ecosystem types, mapping, MAES

P03_02

Aphid pest species on *Chrysanthemum morifolium* Ramat. in Bulgaria

Mariya Yovkova^{1,2}, Aneliya Pencheva³

¹Institute of Ornamental Plants, Sofia1222, Sofia, Negovan, Bulgaria

²Institute of Biodiversity and Ecosystem Research, Bulgarian Academy of Sciences, 2 Gagarin Street, 1113 Sofia, Bulgaria

³University of Forestry, 10 Kliment Ohridski Blvd, Sofia 1756, Bulgaria

Aim: The objective of the present study was to investigate the aphid pest species on *Chrysanthemum morifolium* Ramat. and the effect of the aphid infestations on host plants.

Material and methods: Investigations on the species composition of aphids on *C. morifolium* in more than 20 greenhouses and garden centers in Bulgaria were observed. The present study is conducted in a period of over 10 years, from 2008 to 2018. The observed greenhouses and garden centers are located in the regions of Sofia, Plovdiv, Smolyan, Pavlikeni, Ruse, Varna and Burgas.

Results and discussion: Six different aphid species were detected during the observations. They are *Aphis fabae* Scopoli, 1763, *A. gossypii* Glover, 1877, *Aulacorthum circumflexum* (Buckton, 1876), *A. solani* (Kaltenbach, 1843), *Macrosiphoniella sanborni* (Gillette, 1908) and *Myzus persicae* (Sulzer, 1776). All of the identified species are polyphagous, except *M. sanborni*. The most important, widespread and harmful aphid pest species on *C. morifolium* was *M. sanborni* detected in greenhouses all year round. The damages were expressed in deformations of stems, leaves and flowers. Due to the heavy aphid infestations the flowers cannot open normally and have low trade quality.

Conclusion: The most important aphid pest on *C. morifolium* is *M. sanborni*. It causes damages and decreases the trade quality of the grown plants and cut flowers. It was found all year round in the investigated greenhouses. Less importance in *Chrysanthemum* cultivation, have all other detected species, which were sporadically founded.

Acknowledgements: We would like to thank to Assoc. Professor Dr. Diana Nencheva for the identification of *Chrysanthemum* plants.

Keywords: *Aphididae*, aphids, *Chrysanthemum*, mums, pests

P03_03

Preliminary results of the eutrophication assessment of the water bodies in the Lower Danube floodplain using remote sensing images and *in-situ* verification

Stefan Kazakov¹, Vulko Biserkov¹, Luchezar Pehlivanov¹, Stoyan Nedkov²

¹Institute of Biodiversity and Ecosystem Research, Bulgarian Academy of Sciences

²National Institute of Geophysics, Geodesy and Geography, Bulgarian Academy of Sciences

Aim: Although the newest satellite images provide more and more sensitive and precise data about the aquatic characteristics of the inland waters like chlorophyll *a* and turbidity, the retrieval of reliable absolute values still requires an additional process of calibration or *in-situ* verification. The aim of the study is to provide a reliable scale for the eutrophication assessment and monitoring of water bodies in the Lower Danube floodplain using Sentinel 2 images.

Material and methods: Freely available Sentinel 2 MSI images were used for chlorophyll *a* and turbidity estimation using built in algorithms of the SeNtinel Application Platform (SNAP). In order of verification, the remote assessment *in-situ* measurements of the same indicators were carried out following ISO standards.

Results: *In situ* data showed high variance of chlorophyll *a* concentration indicating from 6.12 to 212 mg/m³, while the Secchi depth ranged from 0.15 to 2.0 m. The highest chlorophyll *a* concentration was measured in an isolated temporary pool, while the lowest one in a water body permanently connected to the Danube. Data from the satellite images showed distinctive changes in chlorophyll *a* concentration of the permanent water bodies like Srebarna (over 100 ha open water) in temporal and spatial scale. Other floodplain lake - Malak Preslavets met the minimum area of open water (24 ha) for remote sensing observation, while the temporary pools experienced difficulties in satellite data reliability.

Conclusion: Srebarna and Malak Preslavets emerged as reliable objects of investigation. In further studies on smaller water bodies the higher dynamic of their hydrology, vegetation and weather conditions should be considered.

Acknowledgements: The study was funded by the Program for Support of Young Researchers and PhD Students at the Bulgarian Academy of Sciences (Grant no. 17-107/28.07.2017).

Keywords: eutrophication, Sentinel data, remote sensing, Lower Danube

P03_04

Weed species diversity and community composition in organic potato field

Iliana Gerasimova, Totka Mitova

ISSAPP “N. Poushkarov”, Sofia, 7 Shosse Bankya Str.

Weeds are one of the greatest limiting factors to efficient organic crop production. At the same time weeds in agricultural lands are the component of biological diversity in agricultural systems (agrobiodiversity) and have a very important role for supporting the ecological biodiversity sapling different ecosystems services. Scientific investigations show that organic agricultural practices promote biodiversity in agrosystems.

Aim: The goal of the experiment is to define the influence of cover crops for green manure on the dynamics of weed infestation during the potato growth season (*Solanum tuberosum*, L.) in organic cropping system.

Material and methods: All crops in three field crop rotation were grown according to organic standards. Weed density and abundance, weed species richness, weed communities composition and similarity were assessed during two years. Different ecological indexes (Shannon' index of biodiversity (H'), evenness (J'), Simpson dominance index, Sørensen similarity index were applied.

Results: The results show that all analyzed weed parameters were characterized with high dynamics during the potato growth seasons. The biological development of weeds in the cover crop before incorporation to the potato field was limited. At the harvesting stage of potato the weed density is very low. The share of the problematic perennial weeds decreased by applying the leguminous and grass mixture for green manure in potato field.

Conclusion: According to the obtained data a cover crop for green manure and soil tillage in the organic potato field are effective practices for controlling the weed infestation with annual and perennial weeds. Changing two phytocenoses (cover crop and potato) during growing period influenced the weed community parameters, assessed by ecological indexes.

Keywords: organic potato, green manure, weed community, ecological indexes

THEMATIC SESSION IV

LANDSCAPE ECOLOGY

PL04_01

Academician Anastas Ishirkov's contribution to the development of biogeography in Bulgaria (on the occasion of 150 years from his birth)

Assen Assenov

Sofia University "St. Kliment Ohridski", Faculty of Geology and Geography, 15 Tzar Osvooboditel Blvd., 1504 Sofia, Bulgaria

Aim: The main purpose of the paper is to analyze the scientific work of academician Anastas Ishirkov and to highlight his importance as a founder of biogeography in Bulgaria.

Material and methods: Using the concept of the classic German geography, which includes three main sections - physical geography, anthropogeography and biogeography, an analysis of the biogeographic component of academician Anastas Ishirkov's scientific work is made.

Main results: Academician Ishirkov, as a teacher in Varna, in 1892 was sent to specialize in Leipzig where he attended Friedrich Ratzel's lectures. He was undoubtedly influenced by Ratzel's geographic determinism, which he further developed, following the ideas of French philosophers of the 18th century. In his two-volume anthropogeography, under the influence of Darwin's evolutionary theory, Ratzel (1882) considered anthropogeography as part of biogeography. In August 1895, Ishirkov defended his doctoral dissertation on "South Bulgaria" with Professor Ratzel. Ishirkov has laid the foundations of many of the geographic disciplines at Sofia University, including biogeography.

Conclusion: When Ishirkov developed his scientific work, anthropogeography was seen as a part of biogeography and, despite the later separation of anthropogeography, it remained a human science. Contemporary studies have evidenced very popular researches, applying an ecological and evolutionary approach, such as that of C. B. Cox, I. N. Healey and P. D. Moore, whose "Biogeography" book (2010) has undergone eight editions.

Keywords: biogeography, anthropogeography, evolutionary theory

L04_01

First attempt of a hierarchical classification of benthic seascape units in the Bulgarian Black Sea

Iliyan Kotsev

Prof. Fridtjof Nansen Institute of Oceanology, Bulgarian Academy of Sciences

Every seascape-oriented investigation of the submarine environment is normally accompanied by the application of a hierarchical classification scheme aimed at logical subordination of the identified benthic complexes. Such an approach should be consistent, while the resulting classification system - easy to use, open for updates and inclusions in order to facilitate its application or adaptation for further research.

Study area: The present study encompasses the submarine coastal slope between Bolata Cove to the north and Cape Emine to the south (Bulgarian Black Sea).

Aim: To investigate the contemporary seascape pattern of the study area, differentiate the existent seascape units and subsequently group them into higher seascape ranks.

Material and methods: Various archive data sets and scanned hard-copy maps (e.g., of the seabed substrates, seafloor geomorphology, bathymetry, benthic biota, etc.) were integrated and analyzed in GIS in order to generate polygons representing seascape units from the lowest hierarchical rank, i.e., *seascape kind*. They were further grouped into *genera*, *types*, and *classes*, by applying a pertinent set of criteria on every superior level of the suggested classification scheme.

Results: A total of 24 seascape kinds were differentiated and subsequently grouped into 5 genera, 3 types, and 1 class.

Conclusion: The seascape pattern of the study area is largely predisposed by the geological settings and geomorphic properties of the local seabed environment, which largely resemble those of the contiguous coastline sectors. The main actors for the contemporary seascape structure, dynamics and evolution are the present-day coastal geo- and morphodynamical processes.

Keywords: seascape ecology, seascape analysis, coastal seascapes, seascape structure, seabed components, Bulgarian Black Sea coastal zone

L04_02

Lazarus taxa in Animalia kingdom

Borislav Grigorov, Assen Assenov

Sofia University “St. Kliment Ohridski”, Faculty of Geology and Geography, 15 Tzar Osvoboditel Blvd., 1504 Sofia, Bulgaria

Aim: The main aim of the current research is to investigate the presence of Lazarus taxa in the animal kingdom during the last ten years, following the official information, published by the IUCN Red List.

Material and methods: The overview study is based on cameral research and the main focus is the Lazarus effect, occurring in the biological kingdom Animalia. The comparative method is applied in order to gain a broader view of the problem. The difference between the Lazarus taxa and the Zombie taxa, Elvis taxa and Ghost taxa is clarified for more explicitness. Some main discoveries of animal taxa are discussed, which is used as a basis for the present investigation.

Results: The category of 53 species in Red List was changed for the last ten years from EX – Extinct to any other category, proving their rediscovery. More than a half of the taxa are fish (53%), followed by Mollusca (15%) and Amphibia (10%). Most of the species are moved to the CR – Critically Endangered category, followed by those, included in the DD – Data Deficient category. More comprehensive information is provided for some of the Lazarus taxa, aiming for more in-depth understanding.

Conclusion: The Lazarus effect, leading to the rediscovery of species that were thought to be lost forever, is occurring even today. It is a major scientific achievement to find again a taxon, considered dead. However, we should focus our immediate efforts on the conservation of those species that are already a part of the biosphere, rather than hoping for the miraculous resurrection of the Lazarus taxa.

Keywords: Lazarus effect, Lazarus taxa, Animalia, IUCN Red List

P04_01

Radiological monitoring of drinking waters in Bulgaria

Radoslava Lazarova, Ivanka Yordanova, Donka Staneva

„N. Poushkarov” Institute of Soil Science, Agrotechnologies and Plant Protection, BG – 1080 Sofia, Bulgaria

Aim: The aim of the present study was to determine the uranium concentration and the gross alpha activity in water samples from different regions in Bulgaria as a part of the overall water quality monitoring in the country.

Material and methods: 793 water samples were analyzed in the period 01.01.2014 - 31.12.2016.

A spectrophotometric method developed and validated by the Testing Laboratory of Radioecology and Radioisotopic Research at the ISSAPP "N.Pushkarov”, based on the principle of formation of uranium–arsenazo-III complex, was used for the uranium determination.

The gross alpha activity was measured by low background counting of the dry water residue deposited on a planchet after a method developed in accordance with ISO 9696/2010.

Results: Relatively high uranium concentration was determined in some of the samples from the regions of Burgas, Plovdiv, Rakovski, Haskovo and Velingrad and gross alpha activity in the regions of Burgas, Plovdiv, Rakovski, Haskovo, Velingrad, Petrich and Dobrich. The percentage of the samples with relatively high values of natural uranium and gross alpha activity was calculated on annual basis.

Conclusion: From the results obtained, the following conclusions could be made.

In the majority of the samples the values of the radiological indicators under study do not exceed the reference ones.

An increase in the percentage of the samples with high values of natural uranium and gross alpha activity is observed in 2016 when more conservative reference values have already entered into force since 28 November 2015. The relatively high uranium concentration and gross alpha activity are probably due to the specific hydro-geological characteristics of the regions where they were observed as well as due to the closed uranium mining near them.

Keywords: uranium, gross alpha activity, radiological monitoring

THEMATIC SESSION V ECOLOGY AND EDUCATION

PL05_01

Successful implementation of project-based learning in higher education: An example from ecology

Kamelia Yotovska, Victoria Necheva

Sofia University “St. Kliment Ohridski”, Faculty of Biology, Department of Biology Education

In recent years, project-based learning has been successfully applied at the Faculty of Biology of Sofia University "St. Kl. Ohridski". The purpose of this article is to present a theoretically grounded and well-tested technology for project-based training at the higher school. The role of this training, the main types of project-based learning and the requirements for its proper conduction in the higher education system are structured.

A specially developed Criteria Assessment System is described. Project-based training, which was conducted during the academic year 2017-2018 within the discipline of Methodology and Technique of the School Biology

Experiment with 4-year students, from the specialties Biology and Chemistry, Geography and Biology and Biology and English, has been elaborated. The methods used are theoretical analysis and synthesis, SWOT analysis and poll.

The increased tendency to acquire applied knowledge and skills from students during their studies in higher schools requires the demand for practically oriented pedagogical technologies. By applying the idea of learning by doing students face specific problems to solve, the process of acquiring scientific approach in researching and mastering specific methods of analysis is assisted.

Putting the students - future teachers at the center of learning makes them active subjects in the learning process and contributes for their personal progress. The use of these methods is appropriate and is also applied successfully in the traditional classroom. Through the project-based training, the relationship "duration of activity - durability of knowledge" is realized; and a connection between subject areas and elements of real life is implemented.

Project-based learning motivates students by engaging them in the process of their own learning; gives them the opportunity to integrate knowledge from different fields of science and apply them to the development of a specific educational project. Thanks to this, students are active participants and partners in the learning process.

Keywords: project-based learning, learning by doing, higher education, pre-service teachers

L05_01

The school education – a condition to form positive attitude towards environment or ecology

Ekaterina Yovcheva, Veneta Kirkovska

Primary school “Kiril Hristov”, Stara Zagora

Recently, in the past years, educational policy marks the outlines of real changes towards environment in the school education. The aim of the school strategy for ecological education is enriching the ecological knowledge and skills of the students. We look for the actuality of the discussed problem in the realization of the Strategy for Ecological (Ecofriendly) education in school as a new component for personal development of the learners.

Keywords: ecological education, environment, eco-friendly culture, ecofriendly attitude

P05_01

Innovative Integrated Training in Healing Plants Business – TBP

Anna Kujumdzieva, Trayana Nedeva, Aleksander Savov, Tereza Toncheva

Sofia University “St. Kliment Ohridski”

Aim: the aim of TBP project is to offer an innovative educational system to farming trainers, advisors, and farmers to fill the training gap in the cultivation and processing of Medicinal & Aromatic Plants (MAP), recognized as one of the most profitable economic sectors.

Material and methods: desk research and analysis of findings; development of a virtual training tool; design and development of training material; methods for performance of distance, part-time and modular training modes.

Results: The TBP project exploits new approach for shifting of existing lack of training to learner-centered one. It offers innovative and competence-based approach of training in: organic culture of MAP, quality certification of final products, business entrepreneurship in the MAP sector, and medical use of MAP. The training is organized through the development of innovative ICT based curriculum, teaching methodology and virtual tools based on Open Learning Management system while enhancing an integrated approach and presenting the best practices on training by implementing the EQF principles. The virtual tools comprise: interactive web platform (<http://businessplants.eu>), e-learning & training material, open learning recourses, and online MAP encyclopedia. Its exploitation contributes to a successful and sustainable development of the MAP sector since specialized knowledge & skills for cultivation and handling procedures, necessary to certify the quality of the end product are provided.

Conclusion: knowledge of the therapeutic value and possible applications of MAP creates new jobs and provides additional income to project end-users. The TBP project produces an added value, since working power mobility and sharing of common experience ensure durable effect and practical application.

Acknowledgements: The TBP project 2016-1-EL01-KA202-023491 is funded by ERASMUS + programme of EC.

Keywords: MAP, ICT, e-learning, EQF

P05_02

Blended learning in bioinformatics – the SMEs instrument for biotech innovations – BIOTECH – GO

Anna Kujumdzieva, Trayana Nedeva, Aleksander Savov, Tereza Toncheva

Sofia University “St. Kliment Ohridski”

Aim: the aim of the BIOTECH-GO project is to launch an innovative system for VET professionals to improve their workplace basic skills by providing training in bioinformatics - an interdisciplinary field of science, which develops methods and software tools for storing, retrieving, organizing and analyzing biological data.

Material and methods: desk research and analysis of findings; methods for on- and off-line training delivery; creation of professional profiles and corresponding competences, definition of learning outcomes.

Results: The BIOTECH-GO project builds a system for structural description of work-related competences. It introduces an innovative competence-based model for VET professionals, based on the European Qualifications Framework (EQF) and the European Credit System for Vocational Education and Training (ECVET). The application of the “Education for Sustainable Development (ESD)” concept of BIOTECH-GO project offers new training opportunities for trainers through personalized learning paths using individual preferences and job requirements improving in this way their prospects for entrepreneurship. Applying a new learner-centered approach, it improves training in bioinformatics for VET professionals and supports the new, proactive role of trainers and tutors. Knowledge, skills and competence update of VET specialists further contributes to the advancement of biology research in Biotech SMEs through bioinformatics tools application; provides advanced bioinformatics training to SMEs personnel at all levels, from technicians to independent investigators; encourages dissemination of cutting-edge technologies to industry.

Conclusion: The project initiative helps job seekers, employers, regulators and policy makers to participate in the shift of the EU labor market towards smarter and greener jobs for sustainable European economy.

Acknowledgements: The BIOTECH-GO project 2016-1-BG01-KA202-023686 is funded by ERASMUS + programme of EC.

Keywords: bioinformatics, blended learning, VET, EQF, ECVET

THEMATIC SESSION VI OTHER RELATED TOPICS

P06_01

Super-critical carbon dioxide as effective eco-friendly solvent for the extraction of purslane (*Portulaca oleracea* L.)

Sabina Taneva, Svetlana Momchilova, Angel Konakchiev, Vladimir Dimitrov

Institute of Organic Chemistry with Centre of Phytochemistry, Bulgarian Academy of Sciences

The aim of this study was to evaluate the extraction of purslane with super-critical CO₂ concerning the yield at different conditions (temperature, pressure) and to compare the results to that of three common classic methods for plant extraction (of Folch, of Bligh and Dyer, and with hexane in Soxhlet apparatus).

Material and methods: Purslane (*Portulaca oleracea* L.) was collected in August 2017 in the region of Novo Zhelezare (near Plovdiv). Stalks and leaves were finely chopped, dried at room temperature in shadow, then ground finely and subjected to extraction. Portions of 800 g were used for the super-critical CO₂ experiments at different temperatures (55 and 80°C) and pressures (100, 255 and 400 bar). 15 g were extracted by the method of Folch (with chloroform-methanol 2:1 mixture), of Bligh and Dyer (with chloroform-methanol 1:2 mixture) and in Soxhlet extractor (with hexane, 6 hours). The yield was determined gravimetrically.






Results: The experiments with super-critical CO₂ revealed that increased temperature and pressure improved the yield. On the other hand, the three classic methods applied ensured yield of about 4% which was lower than that of 6.2% achieved by super-critical CO₂ at 80°C and pressure of 400 bar.











Conclusion: Extraction of purslane with super-critical CO₂ ensures higher yield than that of the common methods, which use solvents as hexane and chloroform-methanol mixtures. Moreover, the super-critical CO₂ is undoubtedly the preferred solvent being non-toxic, non-corrosive, non-flammable and cheap.






Acknowledgements: Financial support provided by the Program for supporting of young scientists and PhD students at the Bulgarian Academy of Sciences – 2017, Project ДФНП 17-60, is gratefully acknowledged. The equipment for super-critical CO₂ extraction is purchased by Grant BG161PO003-1.2.04-0007-C0001 under OP Development of the Competitiveness of the Bulgarian Economy 2007-2013.








Keywords: purslane, *Portulaca oleracea*, super-critical CO₂ extraction

INDEX

Name	Code	E-mail	Student/ PhD student	Page
Ahmed, Atidzhe	P01_01, P02_01, P02_02	atidje_dj@abv.bg		21, 39, 40
Aleksova, Michaela	PL02_02, P02_08			32, 44
Anev, Svetoslav	P02_06			42
Aneva, Ina	P01_16			31
Angelova, Lilia	L02_01	lilia_stoickova@abv.bg		33
Angelova, Tsveta	L02_04, L02_05	angelova_ts@abv.bg		35, 36
Apostolova, Emilia	L02_02			34
Asenov, Asen	P01_04	asenasenov71@yahoo.com		23
Assenov, Assen	PL04_01, L04_02	asseni.assenov@gmail.com		51, 52
Assyov, Boris	P01_02, P01_12, P01_14	contact@boletales.com		22, 28, 29
Babenko, Lilia	L02_01			33
Bakardzhiev, Kamen	P01_14			29
Bancheva, Svetlana	L01_10, P01_11	sbancheva@yahoo.com		18, 27
Banov, Krasen	P02_07	krasen.banov@gmail.com		43
Bardarov, Krum	L01_13			20
Bardarov, Ventzislav	L01_13	vbardarov@abv.bg		20
Berkov, Strahil	L02_07			38
Berov, Dimitar	PL03_02	dimitar.berov@gmail.com		47
Biserkov, Valko	PL03_02, P03_03	v@biserkov.com		47, 49
Bogutskaya, Nina	L01_02	Nina.Bogutskaya@NHM- WIEN.AC.AT		13
Boteva, Silvena	PL02_02, P02_08			32, 44
Bozok, Fuat	P01_02			22
Bratanova-Doncheva, Svetla	P03_01	sbrat@abv.bg		48
Chaneva, Ganka	P02_06	gchaneva@abv.bg		42
Chankova, Stephka	L02_04, L02_05, L02_06, L02_07	stephanie.chankova@yahoo.com		35, 36, 37, 38
Chavgov, Vasil	P02_06			42
Dalakchieva, Svetla	P01_13	s.dalakchieva@gmail.com		29
Danov, Danail	L01_05			15
Danova, Kalina	P02_05	κ_danova@abv.bg		42
Daskalov, Georgi	PL03_02			47
Delcheva, Malina	L01_10, P01_11	malina.delcheva@gmail.com		18, 27
Denchev, Cvetomir M.	P01_03	cmdenchev@yahoo.co.uk		23
Denchev, Teodor T.	P01_03			23
Dimitrov, Martin	L02_05			36
Dimitrov, Vladimir	P06_01			56
Dimitrova, Velmira	L02_02	pmmaster2001@yahoo.com		34
Diripasko, Oleg	L01_02			13

Dobrev, Petre	P02_05			42
Doncheva, Snezhana	L02_02			34
Doycheva, Iva	L01_09			17
Elshishka, Milka	PL01_03, P01_17	melshishka@gmail.com		12, 31
Ganeva, Anna				
Gecheva, Gana	P01_10, L03_01			27, 47
Genov, Peter	P01_01			21
Georgiev, Stoyan	L01_08			17
Georgieva, Galia	L01_14	tsambi@abv.bg		21
Georgieva Ognyanova-Rumenova, Nadya	P01_09			26
Gerasimova, Iliana	P03_04	ilianich_ilieva@abv.bg		50
Gesheva, Emilia	L02_02			34
Gevrenova, Reneta	L01_13			20
Glogov, Plamen	L01_01	pglogov@abv.bg		13
Gospodinov, Galin	P01_07	rpnmgospodinov@gmail.com		25
Grigorov, Borislav	L01_08, L04_02	borislav.g.grigorov@gmail.com		17, 52
Gumus, Media	L01_08	med_i@abv.bg		17
Gyosheva, Melanya	P01_07			25
Hristov, Peter	L01_03, L02_03			14, 35
Ihtimanska, Mila	P02_04	mila.ihtimanska@gmail.com		41
Iliev, Ivan	L02_05			36
Ilieva, Teodora	L03_01	teodoraailieva@mail.bg		47
Ilkova, Julia	P02_04			41
Ionkova, Iliana	PL02_01			32
Isheva, Tsvetelina	P01_09	tsvetelina.isheva@abv.bg		26
Ivanova, Albena	L02_01			33
Ivanova, Nevena	PL03_01	larus@abv.bg, nevena759344@gmail.com		46
Jordanova, Michaela	P01_16			31
Kachamakova, Maria	L01_12	maria.n.kachamakova@gmail.com		19
Karamfilov, Ventsislav	PL03_02	ventzi.karamfilov@gmail.com		47
Katrandzhiev, Kostadin	P03_01	kmkatrandjiev@gmail.com		48
Kazakov, Stefan	P01_08, P03_03	sakazak@mail.bg		25, 49
Kenarova, Anelia	PL02_02, P02_08			32, 44
Kerakova, Maria	L01_06, P01_08	mariqkerakova@abv.bg		15, 25
Kirkovska, Veneta	L05_01			54
Klayn, Stephania	PL03_02			47
Konakchiev, Angel	P06_01			56
Kosakivska, Irina	L02_01			33
Koshev, Yordan	L01_12			19
Kostadinov, Kostadin	L02_05			36
Kotsev, Iliyan	L04_01	ikotsev@io-bas.bg		51
Kozuharova, Ekaterina	PL01_02, P01_16, PL02_01	ina_kozuharova@yahoo.co.uk		12, 31, 32
Kozuharova, Elisaveta	L01_05	ellie_kozuharova@abv.bg		15
Krasteva, Ilina	PL02_01			32

Krumova, Sashka	P02_03			40
Kujumdzieva, Anna	P02_09, P05_01, P05_02			44, 54, 55
Lambevska-Hristova, Aneta	P01_07			25
Lazarova, Radoslava	P04_01	radoslava2005@abv.bg, ilrri.poushkarov@gmail.com		53
Lazarova, Stela	PL01_03, P01_17			12, 31
Manasiev, Jordan	P02_08			44
Mardari, Constantin	P01_10			27
Marinov, Miroslav	L01_03	maff97@abv.bg		14
Marinova, Adriana	P02_06			42
Markov, Georgi	P02_01, P02_02	georgimar@gmail.com		39, 40
Markovska, Yuliana	L02_02			34
Maslenkova, Liliana	L02_01			33
Melika, George	P01_15			30
Michailova, Paraskeva	P02_04			41
Mincheva, Irena	P01_11, P01_16	irena_mincheva@yahoo.co.uk		27, 31
Miteva, Daniela	L02_06	daniela.miteva@abv.bg		37
Mitova, Totka	P03_04			50
Mitrovska, Zhana	L02_04, L02_05, L02_06, L02_07			35, 36, 37, 38
Mladenov, Aleksandar	PL01_03			12
Momchilova, Albena	L02_01			33
Momchilova, Svetlana	L02_01, P06_01			33, 56
Mossa, Abdel-Tawab H.	L02_05			36
Motyka, Vaclav	P02_05	Motyka@ueb.cas.cz		42
Natcheva, Rayna	L01_08, P01_07			17, 25
Nazurov, Momchil	P01_10	jackinblack24@gmail.com		27
Necheva, Victoria	PL05_01	v.necheva@abv.bg		53
Nedeva, Trayana	P05_01, P05_02	nedeva.ts@abv.bg		54, 55
Nedkov, Stoyan	P03_01, P03_03			48, 49
Neov, Boyko	L02_03			35
Nikolova, Milena	L02_07			38
Oleskina, Yuliya	EP02_01			45
Parvanova, Petya	L02_05, L02_07	petq_parvanova@abv.bg		36, 38
Pavlova, Dolja	L01_01			13
Pehlivanov, Luhezhar	P01_08, P03_03			25, 49
Pencheva, Aneliya	P03_02			49
Peneva, Vlada K.	PL01_03, P01_17	vpeneva@ecolab.bas.bg		12, 31
Petrov, Stefan	L01_09			17
Petrova, Antoaneta	P01_13, P01_14	petrovabotgar1@abv.bg		29, 29
Petrova, Detelina	P02_06			42
Petrova, Galya	L01_09	galiaty@abv.bg		17
Petrova, Nia	P02_03	petrova_nia@yahoo.com		40
Petrova, Ventsislava	P02_09	v.petrova@biofac.uni-sofia.bg		44
Popatanasov, Andrey	L01_11	and_atanasov@abv.bg		19
Popov, Mladen	P02_08			44

Popova, Elitsa	P01_01	elitsa.d.popova@gmail.com		21
Radeva, Galina	PL02_02, P02_08	galrad@abv.bg		33, 44
Radoslavov, Georgi	L01_03, P01_17, L02_03			14, 31, 35
Rimcheska, Biljana	L01_07	rimceska@gmail.com		16
Savov, Aleksander	P05_01, P05_02			54, 55
Sharapova, Lubov	EP02_01			45
Shivarov, Veselin V.	L01_08			17
Shkondrov, Aleksandar	PL02_01			32
Sidjimova, Boriana	L02_07			38
Simeonova, Valeria	L02_04			35
Sirakova, Daniela	L02_03			35
Slavova, Monica	P01_12			28
Soufi, Rabia	L01_06	rsoufi@gmail.com		15
Stanilova, Marina	PL01_01, L01_09, P01_05, P01_11	marina.stanilova@gmail.com		11, 17, 24, 27
Staneva, Donka	P04_01	donastaneva@abv.bg		53
Stateva, Stanislava	P01_06	stanislava.stateva@gmail.com		24
Stefanov, Martin	L02_02	martin_12.1989@abv.bg		34
Stefanov, Tihomir	L01_02, L01_04	tishos@gmail.com		13, 14
Stepanov, Ivan	P01_01			21
Stoichev, Svetozar	P02_03	svetlio85@abv.bg		40
Stojanova, Anelia	P01_15	stanelia@uni-plovdiv.bg		30
Stoyanov, Stoyan	L01_09			17
Taneva, Sabina	L02_01, P06_01	sabu_09@abv.bg		33, 56
Taneva, Stefka	P02_03			40
Taşkın, Hatira	P01_02			22
Teofanova, Denitsa	L01_03			14
Todev, Ivan	P01_01			21
Todinova, Svetla	P02_03			40
Todorov, Ivaylo	P01_15	i.toddorov@abv.bg		30
Todorova, Teodora	L02_05	tedi_todorova@yahoo.com		36
Tomova, Anna	P02_09	aderekova@yahoo.com		44
Toncheva, Tereza	P05_01, P05_02			54, 55
Traykova, Boryanka	P01_05	borianka_traikova@abv.bg		24
Tzvetkova, Nikolina	P02_06			42
Uzunov, Yordan	L01_14	uzunov@ecolab.bas.bg		21
Uzundzhalieva, Katya	P01_06			24
Varadinova, Emilia	L01_06, P01_08			15, 25
Vassilev, Kiril	L01_08, P01_10, L03_01	kiril5914@abv.bg		17, 27, 47
Vassilev, Rosen	P01_14			29
Velev, Nikolay	L01_08			17
Vidinova, Yanka	L01_07			16
Yankova, Iskra	L02_03	s.scardica@gmail.com		35
Yordanova, Ivanka	P04_01			53
Yotovska, Kamelia	PL05_01			53

Yotsova, Ekaterina	L02_02	ekaterina_yotsova@abv.bg		34
Yovcheva, Ekaterina	L05_01	piperska@abv.bg		54
Yovkova, Mariya	P03_02	mariya_yovkova@abv.bg		49
Yurina, Nadejda P.	L02_06, EP02_01	nyurina@inbi.ras.ru		37, 45
Zafirov, Nikolay	P02_07	niki.zafirov@gmail.com		43
Zheleva-Dimitrova, Dimitrina	L01_13			20
Zhiponova, Miroslava	P02_06			42
Zlatkov, Boyan	P01_15	bzlatkov@gmail.com		30