

Macedonian Ecological Society

**5th CONGRESS OF ECOLOGISTS
OF THE REPUBLIC OF MACEDONIA
WITH INTERNATIONAL PARTICIPATION**

ABSTRACT BOOK

Ohrid, Macedonia 19th - 22nd October 2016

Издавач: **Македонско еколошко друштво**

Институт за биологија

Природно-математички факултет - Скопје

П. фах 162, 1000 Скопје

Цитирање:

Книга на апстракти, V Конгрес на еколозите на
Македонија со меѓународно учество. Охрид,
19-22.10.2016. Македонско еколошко друштво,
Скопје, 2016

Publisher: **Macedonian Ecological Society**

Institute of Biology

Faculty of Natural Sciences

P.O. Box 162, 1000 Skopje, Macedonia

Citation:

Abstract book, V Congress of Ecologists of the
Republic of Macedonia with International Participa-
tion. Ohrid, 19-22.10.2016. Macedonian Ecological
Society, Skopje, 2016

CIP - Каталогизација во публикација

Национална и универзитетска библиотека "Св. Климент Охридски", Скопје

502/504(062)(048.3)

CONGRESS of ecologists of the Republic of Macedonia with international participation
(5 ; 2016 ; Ohrid)

Abstract book / 5th Congress of ecologists of the Republic of Macedonia with international participation, Ohrid, Macedonia 19th - 22nd October 2016 = Книга на апстракти / [V Конгрес на еколозите на Македонија со меѓународно учество. Охрид, 19.-22.10.2016]. - Скопје : Македонско еколошко друштво = Skopje : Macedonian Ecological Society, 2016. - 213 стр. ; 25 см

Текст напоредно на мак. и англ. јазик

ISBN 978-9989-648-36-6

I. Конгрес на еколозите на Македонија со меѓународно учество (5 ; 2016 ; Охрид) види Congress of ecologists of the Republic of Macedonia with international participation (5 ; 2016 ; Ohrid)

а) Екологија - Собири - Апстракти

COBISS.MK-ID 101812746

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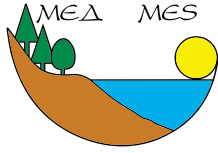
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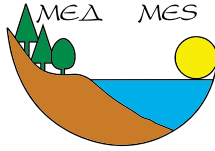
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Section 1

PLENARY LECTURES



Conservation of Reptiles in the central Balkans – *de jure & de facto*

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There is no doubt that populations of reptiles are globally declining at large scale. Recent wide-ranging analysis of conservation statuses and threats of reptiles of the world revealed that nearly 20% of the species are threatened, while for another 20% data are inexistent. Unambiguously, only long-term population studies could prove global declines of different reptilian taxa, but they are logistically difficult, costly, time consuming and therefore very rare in herpetology. Consequently, global extinction risk of many reptile species is set primarily upon their distribution data, or/and expert opinions, without a proper support from population studies. As concerned to reptile conservation, the situation in the central Balkans (Serbia, Macedonia and Montenegro) is even worse. In all three countries, there is a sharp discrepancy between formal (legal) conservation statuses (*de jure*) and actual (*de facto*) situation in conservation practices of reptiles. Majority of reptile species present in the countries in concern are either protected or strictly protected at national levels: 72 % (23 of 32 species) in Macedonia, 76 % (26 of 34 species) in Montenegro and 83 % (20 of 24 species) in Serbia. However, in some cases, lists were made upon experts (?) opinions, while in other, species with international significance were omitted (e. g. Viperidae in Montenegro). Prerequisite for setting conservation priorities of reptiles are existence of national Red Lists and/or Red Books of Macedonia and Montenegro. This requires publication of detailed distribution of reptiles (in Montenegro), updated analyses of centres of diversity (in Macedonia), and whenever possible, inclusion of population parameters and trends (in all three countries). In this presentation, results of long-term population and conservation studies will be provided, as an example how they could be used in estimation of conservation statuses of internationally important (e. g. *Testudo hermanni*) or neglected (e. g. *Natrix tessellata*) reptile species.

Keywords: herpetofauna, conservation priorities, Red Books, legal protection

Check list of European phytosociological alliances – a new challenge for vegetation science

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Vegetation classification according to the Braun-Blanquet methods is widely used in Europe. After 100 years of classification efforts of European phytosociologists (vegetation scientists); many researches have been done and concepts and names of vegetation units have been proposed, but there was no classification system integrating all these units. This was the reason that the group of researchers of over 30 researchers guided by L. Mucina have evaluated about 10 000 bibliographic sources

and created a comprehensive list of syntaxonomic units till the alliance level appearing all over the Continent, from Canary Islands till Ural and from Cyprus to Greenland. These units were evaluated by experts for their floristic and ecological distinctness, geographical distribution and compliance with nomenclatural code. The list contains 109 classes, 300 orders and 1104 alliances. The aim of the project was to document the classification and to stabilize the concepts and nomenclature of syntaxa also for practical uses such as calibration of habitat classification, standardization of terminology for environment assessment, management and conservation. The present classification system provides a baseline for future development and revision of European syntaxonomy.

Keywords: classification, Europe, phytosociology, systematics, vegetation.

Environmental pollution with heavy metals in the Republic of Macedonia

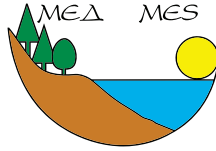
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Anthropogenic environmental changes, associated mainly with chemical pollution, lead to a degradation of the natural human environment. Among all chemical pollutants, trace elements are of a special ecological, biological and health significance. The production of energy and the consumption of natural resources are the main source of trace elements as contaminants. However, agricultural activities, especially the application of fertilisers and pesticides, also contribute significantly to trace metal pollution in the environment. Soil, as a part of the ecosystem, is vital for the survival of mankind which is closely connected to its productivity. Therefore, the surveys of the pollution with heavy metals of soil, waters, sediments, air and food on the whole territory of the Republic of Macedonia were performed. For that purpose various spectrometric (atomic absorption spectrometry, inductively coupled plasma – atomic emission spectrometry, and inductively coupled plasma – mass spectrometry) and radioanalytical (neutron activation analysis) techniques were applied. Air pollution was investigated by the application of moss biomonitoring and dust samples (attic dust and house-hold dust). The pollution with heavy metals in the particular regions was additionally investigated. It was found that the highest pollution is present in the areas with abounded or active mines (Pb, Zn, Cu, As, Sb, Ni, Cr), metallurgical plants (Pb, Zn, Cd, Fe-Ni, Fe-Cr, Fe-Si, Fe, steel) or thermoelectrical power plants. High content of some heavy metals were also found in the areas where their contents usually vary gradually across the geochemical landscape and depend on the geochemistry of the underlying lithology. The distributions of such elements reflect natural processes indicated by the elements that are either rarely or never involved in the industrial processes. The obtained data are statistically processed and spatial distribution maps for each specific element are prepared to give a proper interpretation of the obtained results.

Keywords: heavy metals, pollution, air, soil, water, sediments, Republic of Macedonia



Section 2

**POPULATIONS, COMMUNITIES
AND ECOSYSTEMS**

Carabid beetles diversity at abrupt and successional forest edges

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Forest edges are a type of ecotone or transitional zone between adjacent habitats. They are characterised by unique physical appearance, which could manifest in changes in the abiotic and biotic factors. Carabid beetles were studied at abrupt and successional forest edges, adjacent forest interior and grassland. The study was performed in the Gorski Kotar region (Croatia) in the belt of Dinaric beech-fir forests. Beetles were collected during two consecutive years, using pitfall traps. Edge effect was detected at the carabid beetle assemblage level, i.e. diversity was significantly higher in the forest edges in comparison to the forest interior. Both forest edges acted as filters for open habitat species, while they were a barrier for certain forest specialist. Activity density was significantly higher in the forest interior, due to dense populations of forest generalist. Carabid assemblages in the forest edges showed a high degree of similarity with the assemblages in the forest interior. Soil temperature, soil humidity and canopy openness differed significantly between forest edges and forest interior. Thus, vegetation structure and soil properties may play a key role in determining the spatial pattern of carabid beetles in the forest edges.

Keywords: ecotone, transitional habitats, community analyses, activity density, species richness

Spatial distribution and community structure of ground-beetles (Coleoptera: Carabidae) along altitudinal gradient on Belasitsa Mt., R. Macedonia

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Ground beetle abundance and community characteristics were compared along an altitudinal gradient on Belasitsa Mt. The material was collected monthly during a period April-November 2010, in 14 localities, with pitfall traps. Individual-based rarefaction method was used to compare species richness between the localities and dissimilarities in ground beetles' assemblages between localities were analyzed with cluster analysis. In total 8680 specimens belonging to 38 species, 18 genera, 11 tribes and 7 subfamilies were recorded. *Tapinopterus balcanicus belasicensis* Maran 1933, *Molops rufipes belasicensis* Mlynar 1977 and *Carabus convexus dilatatus* Dejean 1826 were by far the most abundant species. There was clear separation of three groups of habitats based on differences in community structure of ground-beetles: lower altitudinal zones (240-767 m a.s.l.), habitats at higher altitude (847-1385 m a.s.l.) and marked separation of the clear-cut area (1442 m a. s.l.). Higher species asymptotes of rarefaction curves, coupled with significantly higher values of Shannon diversity and

Pielou evenness indices in the localities from lower altitudinal belt clearly indicate that altitude, and consequently related differences in vegetation type along the gradient, affects species composition and richness of ground beetle communities.

Keywords: ground-beetles, ecology, altitude, Belasitsa Mt.

Zoogeographic structure, habitat and humidity preference of ground-beetles (Coleoptera: Carabidae) on Belasitsa Mt.

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Zoogeographical distribution, as well as habitat and humidity preferences of ground-beetles (Coleoptera: Carabidae) are presented in this paper. The material was collected monthly in the period April 2010- November 2010, with pitfall traps from 14 different localities (from the base - 240 m above s.l. to the top -1500 m above s.l. of the mountain) on the north slopes of Belasitsa Mountain. Along altitudinal gradient, the abundance of ground beetles with Subendemic, Locally Endemic, Central-Asiatic, Holarctic and Palearctic distribution significantly increased, unlike South-European and East-Mediterranean ground-beetles which showed significantly lower abundance in localities at higher altitude. Regarding habitat preference, decreased abundance of forest specialists in the beech forest and their absence in the clearcutted area, respectively, is a reliable indicator of certain degree of anthropogenisation and degradation of the beech forest. Increasing altitude and consequently differences of vegetation type (from oak to beech forest and to clearcutted space) lead to significant decrease of mezoxerophilic abundance, and increase of mezohigrophilic and hydrophilic species.

Keywords: Carabidae, zoogeography, habitat and humidity preference, community, altitude

Ecology and distribution of the araneocenosis in the Skopje and Malesh valleys of R. Macedonia

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Data concerning ecology and distribution of Araneae of the Skopje and Malesh valleys in R. Macedonia is presented. 40,77 individuals per trap were collected from six habitats, with pitfall traps in the period of April-August 2014. Their structural characteristics were assessed by using indexes of

richness, diversity, homogeneity and dominance. The structure of the spider community inhabiting Malesh and Skopje valleys differs with highest species richness in the agrarian, and highest relative abundance in the riparian habitat, while the index of dominance reached highest values in the pine forest. Wandering spiders, Lycosidae and Gnaphosidae, were by far the most abundant and species rich families. In general, noticeable differences of the araneocoenosis inhabiting six different habitats were registered, mainly as a result of differences in the altitudes, climate and dominant vegetation types.

Keywords: community structure, Araneae, structural indexes

Diversity and temporal relationships between mammals at feeding stations in Western Rhodope Mountains, Bulgaria

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Supplementary feeding of game species is a widely-spread practice throughout Europe. This leads to concentration of game and non-game species, which develop a coexistence strategy to utilize the resources with the least energy cost. Supplementary feeding practices have both beneficial and negative impacts on numerous species at different ecosystem levels. This brings the need to study the diversity and behavior of these species at the feeding stations which until now have been poorly studied. The objectives of our study were to identify the species visiting the feeding stations and investigate the temporal relationships between the mammals there. 52 camera traps were placed at feeding stations in 7 State Hunting Enterprises in Western Rhodope Mountains, Bulgaria. The captured data was processed and analyzed with a modified version of CameraBase 1.6. A total of 23 species were registered: 14 mammals and 9 birds. According to the estimated Relative Abundance Indices (RAI) the wild boar (*Sus scrofa*), red deer (*Cervus elaphus*) and roe deer (*Capreolus capreolus*) are the most frequently visiting mammals. For each camera trap, the succession of mammal visits was analyzed, as well as the duration of each visit and the time spacing between them. 12 combinations of two or more mammal species recorded at the same time were documented. These include the simultaneous presence of a brown bear *Ursus arctos* (in some cases a female with a cub) and another species like the wild boar and red fox (*Vulpes vulpes*). The animals employ different strategies to utilize the supplementary food and manage to avoid strong competition. Feeding sites provide a unique opportunity to study the behavior of various mammals, including ones with conservation significance.

Keywords: time spacing, activity, competition, brown bear, wild boar

Taxonomic attribution of natural isolates of entomopathogenic fungi in certain regions in Switzerland and their impact in the cockchafer grubs' control

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The grubs of common cockchafer (*Melolontha melolontha*) and forest cockchafer (*Melolontha hippocastani*) are one of the most important insect soil pests. The larvae feed on different plant roots causing serious damages in grasslands and orchards. One of the most successful control methods is the implementation of entomopathogenic fungi as biological control agents. In this study we investigate the natural status and taxonomic determination of the entomopathogenic fungi in field studies of two different regions in Switzerland. The most dispersed of the entomopathogenic fungi were revealed the species of the genus *Metarhizium*, within which *Metarhizium brunneum* was revealed to be the most prominent fungal species. Other isolated strains belonged to species of the genera *Lecanicillium* and *Isaria*. The well-known fungal strain in the biological control of cockchafer grubs, *Beauveria brognartii*, was observed very rarely.

The accurate taxonomic determination with molecular biological methods by amplifying DNA *EF- α* (elongation factor) sequences showed clear differences between the fungal strains within species *M. brunneum*, *M. anisopliae* and *M. robertsii*.

The grade of the pathogenicity of different fungal strains assessed by a bioassay trial against cockchafer grubs, did not show a favourite fungal strain. These experiments have to be repeated in this season. Although the results show several interesting candidates with similar level of performance in biocontrol, as the already commercialized *Beauveria brognartii*.

Keywords: *Melolontha melolontha*, *Metarhizium* sp., *Beauveria brognartii*, *Isaria* sp., *Lecanicillium* sp., biocontrol, fungal strains.

An evaluation of meristic characters in gudgeons (Cyprinidae: Gobioninae) from R. Macedonia

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Gobio and *Romanogobio* are the only native European freshwater cyprinid genera of the subfamily Gobioninae. They comprise a morphologically and ecologically diverse group of fishes, being a subject of many taxonomic studies. In the past in R. Macedonia four subspecies have been established (*G. gobio ohridanus*, *G. gobio balcanicus*, *G. uranoscopos stankoi* and *G. kessleri banarescui*). However,

the recent applications of different species concepts and discarding the category subspecies necessitates a revision of the taxonomic status of the gudgeons, including the ones from the Balkan Peninsula.

The purpose of this study is to reevaluate the morphological characters used in the description of the gudgeons in Macedonia, with focus on the meristic characters. The examined material was taken from the collections of the National Museum and Institute for Animal Science, and from the type locality of the species sensu Dimovski and Grupce. Multiple correspondence analysis (MCA) was used to calculate categorical data in Statistica 7,0 software package.

The results showed that the meristic characters offer a unique phenotypic clue for taxonomic discrimination. Multiple correspondence analysis (MCA) showed an association between qualitative variables, such as scales, fin rays and barbel length, especially between *Gobio* species from Ohrid Lake and Vardar drainage. These results are invaluable for future investigations of the biology and the ecology, as well as for a better understanding of the phylogeny of the gudgeons. Since a species is an important functional term, these results can enable accessing biodiversity and making conservation plans.

Keywords: morphology, multiple correspondence analysis, *Gobio*, meristic counts

Seasonal variation in the body shape of the rotifer *Keratella quadrata* (Müller, 1786) in several Albanian brackish water bodies

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Seasonal/temporal variation in body shape, often termed cyclomorphosis, occurs in several zooplankton groups, including. Such variation is common within the genus *Keratella*, chiefly involving changes in body and spine lengths. Changes in body shape of the rotifer *Keratella quadrata* in three important brackish water bodies (Karvasta, Narta and Butrinti) were recorded over the course of half a year (mid-November 2013 to mid-June 2014). Following our data records body and caudal spine lengths increased from November until the end of February, and then decreased until June. Such changes were not significantly correlated consistently positively or negatively to water temperature, pH, conductivity, food levels or total number of copepod numbers. Following individuals recorded during investigations during the winter season individuals corresponded most closely to the literature descriptions of the group or subspecies *quadrata*, while those present during the spring-summer shows the form, group or variety *dispersa*. The first half of the study period was characterized by water of relatively low temperature, low pH and high conductivity, while, for the second half, temperature and pH increased and conductivity decreased.

Keywords: zooplankton, rotifer, cyclomorphosis, *Keratella*, Butrinti, Karavasta

Differential herbivory on dioecious forest perennial: light-biased, sex-biased or both?

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The omnipresent plant-herbivore interactions and selective pressures imposed by herbivorous insects are among the central themes in plant evolutionary ecology. Light environment can shape these interactions, affecting diversity of plant defences and resulting in differential herbivory. Also, in dioecious plants male and female function are separated, and can differ in susceptibility to herbivores. Differential herbivory in sun and shade has attracted much research interest recently, and growing number of studies obtained different results. However, little is known about how light-mediated changes in defensive and nutritive traits in combination with plant sex affect the degree of herbivore damage. Thus, we investigated the effects of light environment and sex on natural herbivore loads in dioecious forb *Mercurialis perennis*. Plants from shade and sun were analysed with respect to foliar damage (calculated as average percentage of leaf area removed), specific leaf area, carbon-based defensive compounds, nutritional quality and plant size. We explored the role of these traits in predicting the extent of damage inflicted by herbivores, as well as the patterns of sexual dimorphism. Overall, we recorded significant light-differential herbivory. Foliar damage was higher in sunny environment, although the amount of tannins was greater. Specific leaf area was the trait most prominently related to herbivory load, while the effect of leaf nutritional quality was moderate. We conclude that factors underlying risk of consumption were related to plant physical traits and nutritive quality rather than to chemical defenses. The effects of sex on herbivory loads differed markedly between light environments. We recorded contrasting patterns of intersexual differences in total condensed tannins and soluble proteins in sun and shade. We discuss the obtained results in light of current hypotheses on light-biased and sex-biased herbivory and future research needed to fully understand factors underlying the observed herbivory patterns and complex evolutionary interplay among growth, reproduction and defense in plants.

Keywords: folivory; *Mercurialis perennis*; herbivory in sun and shade; defense traits; plant sexual dimorphism

Electron microscopic observations of interlobular interstitium of two salmonids from Ohrid Lake during the reproductive period

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The ultrastructure of interlobular interstitium of Ohrid trout (*Salmo letnica* Kar.) and Ohrid belvica (*Acantholingua ohridana*) during the reproductive period was investigated by transmission

electron microscope observations. The vascularization on the level of interlobular interstitium in the period before spawning is enormously narrowed, brought to minimum, except in the hilus region, where the vascularisation and the cellular elements are remarkably noticed. The cellular components, fibroblasts and myoid elements are rare in the connecting stroma. Rare isolated macrophages can be seen. During the period after the spawning interlobular interstitium is remarkably developed. On the level of interstitium vascularisation is clearly distinct, with visible cellular infiltration. As to the cellular components besides Leydig cells, fibroblasts, collagenous fibrils and myoid elements are remarkably present. The myoid elements are with smooth muscular nature. In their cytoplasm the presence of microfilaments can be noticed.

Keywords: Ohrid trout (*Salmo letnica* Kar.), Ohridska belvica (*Acantholingua ohridana*), testes, interlobular interstitium, ultrastructure.

Biocenological study of arthropod community along altitudinal gradient on Belasitsa Mt. (R. Macedonia)

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Biocenological investigation of arthropods along altitudinal gradient on Belasica Mountain was made. The research was carried out in the period April-November 2010, at 14 sampling sites and pitfall traps were used. During the study altogether 61508 specimens were captured, belonging to 146 taxons, 97 families, 23 orders and 8 classes. The largest share was represented by beetle community (Coleoptera - 110 taxa). Four species and three genera were new for Belasitsa Mountain and for the fauna of R. Macedonia, five rare and four endemic taxons were registered, while five taxons were noted in the International Lists for Biodiversity Protection. Despite arthropod diversity which significantly decreased, the species diversity of Araneae, Orthoptera, Myriapoda, Coleoptera did not change significantly along the altitudinal gradient and consequently with the differences of vegetation type. The average abundance of arthropods in general, as well of Orthoptera and Myriapoda didn't show significant correlation with altitudinal gradient and vegetation type, unlike Araneae and Coleoptera whose abundance was positively correlated with increasing altitude and change of vegetation type (oak-beech forest-clearcutted space).

Keywords: community structure, elevation effects, Belasitsa Mt.

Effects of altitude on distribution and structure of beetle community (Coleoptera) on Belasitsa Mt. (R. Macedonia)

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Spatial distribution and community structure of beetle fauna along altitudinal gradient on Belasitsa Mt. was studied. During the period April-November 2010, adult beetles were collected at 14 localities, by pitfall traps. A total of 12705 specimens belonging to 110 species, 31 subfamilies and 27 families were found. Species richness of beetles did not change significantly along the gradient. The greatest number of species was found in the old oak forest, at 693 m above s.l. and the lowest in beech forest at 1038 m above s.l. The largest share of diversity was represented by ground beetles (38 taxa), followed by Tenebrionidae and Cerambycidae (13 taxa), while other families occurred with a low number of species (1 to 5). Species richness of Ochodaeidae, Orphnidae and Tenebrionidae significantly decreased along the gradient, Leiodids richness was positively correlated with altitude. Despite species richness, the abundance of beetles significantly increased with altitude and consequently with differences of vegetation type. Significantly lowest abundance of beetles was registered in the oak forest at 587 m above s.l., and highest in the beech forest at 1100-1200 m above s.l. The abundance of Ochodaeidae, Orphnidae, Elateridae, Tenebrionidae and Histeridae significantly decreased, unlike Staphylinidae, Leiodidae and Cryptophagidae whose abundance increased along the gradient.

Keywords: Coleoptera, altitudinal gradient, Belasica Mt.

Ecological patterns of water bugs (Hemiptera: Heteroptera) assemblages in karst springs: a case study in central Montenegro

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The composition of water bug communities from 32 springs located in central part of Montenegro was investigated. Twenty-five species were identified, 13 of them are reported as new for Montenegro. The most common species were *Hydrometra stagnorum* and *Velia affinisfilippi* (Gerromorpha). According to environmental characteristics springs were divided into three groups indicating anthropogenic impact on the spring habitats. Springs were divided into four groups according to the composition of water bug communities. There are differences in species richness between these four types of water bug assemblages and among the studied spring types. Results of CCA analysis revealed spring size as the main driver of biotic diversity of aquatic bugs in springs. Our study showed that

community groups of water bugs specified in the biotic classification of spring habitats are much better defined than the assemblages separated in the environmental site classification.

Keywords: water bug, karst springs, Montenegro

Spatial variability in morphology of Y chromosome in bank vole, *Myodes glareolus* (Mammalia, Rodentia) in Serbia

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Although polymorphism of the Y chromosome in bank vole, *Myodes glareolus*, is a phenomenon known for more than 40 years, few studies just pointing to the presence of different types of Y chromosome, have been done since. Total of 113 males have been caught by live traps at 8 localities from Serbia. Chromosome preparations were done from bone marrow and karyotypes were analyzed for the presence of different types of Y chromosome. Two types of Y, metacentric and acrocentric, were detected. Metacentric Y was prevalent (62.83%). Additionally, differential cytogenetic staining was done to obtain further information about differences between noticed types of Y. In order to check if there is any dependence of variation in frequencies of metacentric Y and geographic parameters (latitude and longitude) multiple regression analysis was done. Coefficient of regression was significant ($R^2 = 0.589$; $F_{(2,5)} = 6.021$, $p < 0.046$) but only latitude showed negative significant correlation with frequency of metacentric Y. Its frequency is increasing from North to South in a range from 0.11 to 1.0.

Keywords: polymorphism, metacentric Y, longitude, latitude

Non-traumatic method for individual identification of *Vipera ammodytes*

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This study describes a method for individual identification of *Vipera ammodytes*. Individual identification of the animals is a key factor for clarification of many ecological aspects of the species, such as population density, life histories, home range, etc. Five polygons were chosen along the south-north gradient of the species distribution in Bulgaria. They were explored monthly between 2013 and 2016, during the active period of the animals. The captured animals were marked with a color pen and were photographed. A total of 264 vipers were captured. Forty (15.15%) of them were positively identified at least once. We successfully identified snakes 54 times, excluding daily resightings. The

color mark served for short term identification, as it lasted up to two months during the active period, and up to four months, during the hibernation of the animals. For long term identification, we used the number, shape and arrangement of the scales of the horn, on the tip of the snout (including Rostrale and Suprarostrale), which remains the same throughout the life of the snakes. Due to traumas, the horn can be damaged therefore, for additional certainty of identification we also used the foliosis and the color pattern of the head, as well as the color pattern of the body. The advantage of this method, compared to other widely used methods, such as scale clipping, is that it's non-traumatic for the animals. Its main flaw is that it is quite laborious, taking into account the large number of pictures that must be compared, for the positive identification of the animals.

Keywords: color marking, scale number, color pattern

Some aspects of lizard ecology: habitat preferences, microhabitat selection and spatial niche partition among five sympatric lizards (Reptilia: Squamata: Lacertidae, Scincidae) in North-West Bulgaria

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Lacertids are an important and characteristic part of the European herpeto communities. The family is represented by nine species in Bulgaria. Sympatric occurrence of more than five species is very rare. The studied species were the Green lizard *Lacerta viridis* (Laurenti, 1768), the Wall lizard *Podarcis muralis* (Laurenti, 1768), the Balkan Wall lizard *Podarcis tauricus* (Pallas, 1814), the Meadow lizard *Darevskia praticola* (Eversmann, 1834), which belong to the family of Lacertidae and the single scincid lizard occurring in Bulgaria - the Snake-eyed skink *Ablepharus kitaibelii* Bibron & Bory de Saint-Vincent, 1833 and co-occur in many of their habitats.

The study area is located in the Montana district near Ogosta Dam in North-Western Bulgaria. A total of 773 individual locations were collected, which fall into the seven general types of habitat. To express the diversity of species per habitat type and the diversity of used habitat types by species, the Shannon diversity index was used. In addition, the abundance of species was calculated as frequencies of occurrence per daily searching events and habitat types and compared by species, seasons and years.

The Green lizard was the species with the widest spatial niche. It was found in all of the habitats but it was most common in the grasslands with shrubs, which provide cover places for sunbathing and observation. The rest of the species had narrower spatial niches. The greatest species diversity was found in the grasslands – all of the five species because of the intermediate position among the other habitat types.

All of the studied species displayed specific habitat preferences thus we have found spatial partitioning in the studied reptile community.

Keywords: Herpetofauna, Bulgaria, diversity

Phonotaxis in the orientation of the Danube crested newt *Triturus dobrogicus* (Kiritzescu, 1903)

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The role of sounds as reference cues in the orientation of amphibians has not only been proven for many anurans, but also in a few urodelans that demonstrated phonotaxis based on call recognition. Nevertheless, this aspect of urodelan orientation is still data-deficient and in need of additional research. In this context we tested whether Danube crested newts could discriminate between calls of the edible frog *Pelophylax esculentus* (a species that lives in syntopy with them) and calls of the agile frog *Rana dalmatina* (a more terrestrial species). Our hypothesis was that newts would be more active and would direct their movement to the familiar call, relating it to a suitable habitat. For the experiments we used a circular plastic pool, 80cm in diameter, with sound stimuli played from the outside at 0, 90, 180 and 270 degrees. Movement was recorded with a video camera positioned above the pool and analysed with the Kinovea software (ver. 0.8.15). Results demonstrated that newts did not discriminate between the two calls and a silent control, neither in their movement direction nor in the distance covered. Although the distance covered during tests with *R. dalmatina* calls was, on average, greater than in the other two tests, this was not statistically significant. While our current sample was too small in order to draw any definite conclusions, the study provides preliminary data on the role of anuran calls in the orientation of this species. Based on our findings, we could surmise that the Danube crested newts either 1) lack the ability to use frog calls as reference cues for orientation (being more aquatic, e.g. less migratory than other crested newts) or 2) are more active when presented with an unfamiliar stimulus (exhibiting a preference for the novel call of the agile frog).

Keywords: acoustic stimulus, behavioural response, movement, urodelans.

Reaction of the slow-worms *Anguis fragilis* and *Anguis colchica* to predators' odour in Bulgaria

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The ability of prey to recognize chemical cues of predators has a very important role for its survival in the natural habitats. The slow-worms *Anguis fragilis* and *Anguis colchica* are widespread in Bulgaria. Their main predators are the viper snakes *Vipera ammodytes* and *Vipera berus*. In the present work we tested the reaction of adult and juvenile *A. fragilis* and *A. colchica* towards the vipers' odours. The experiments were carried out in spring-summer period of 2016. The slow-worms were individually placed in small terrariums, and cotton tips, containing the odours of viper snakes were

presented. Differential tongue-flick rates suggested that the odours from the predators were recognized by the slow-worms. Behavioral events such as looking around and hiding were also taken into account. The observations showed that in juveniles the frequency of tongue-flicks towards the odours of *V. ammodytes* and *V. berus* was significantly lower in comparison with that displayed by adults. No species-specific differences between *A. fragilis* and *A. colchica* to the predators' odours were established. These findings suggest that the ability of *A. fragilis* and *A. colchica* to recognize predators' odour is based on individual experience.

Keywords: odour recognition, tongue-flicks, reptilian behaviour, Anguidae

Intraspecific variation in defense against herbivores in an understory forb: exploring spatial and temporal patterns

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When studying plant - herbivore interactions in microevolutionary context, one of the major challenges is to identify which plant traits have the key defensive role and how their relative contributions to plant resistance in natural populations vary in space and time. Theory and empirical evidence show that environmental heterogeneity, both spatial and temporal, affects the diversity of types and levels of plant defenses and, consequently, the outcome of plant - herbivore interactions. In this study, we explored variation in defenses against folivorous insects in natural populations of *Mercurialis perennis* L. (Euphorbiaceae) from Serbia. The model species is a widely distributed forb typically occupying shady understory of temperate forests. Prior to exploring the patterns of intraspecific variation in herbivore loads, we compared methods of assessing the extent of foliar damage. We analysed a set of plant traits representing parameters of plant size ("apparency"), nutritional quality (water and protein content), physical and chemical defenses (secondary metabolites). We found significant variation in overall percentage of foliar damage, both spatial and temporal, and the traits being the best predictors of the risk of consumption differed among the populations. Secondary metabolites (tannins and phenolics) had generally lesser impact than expected. These results are in line with novel studies indicating that contributions of chemical relative to other types of traits to defense against herbivores can be more complex than believed previously.

Keywords: folivory; secondary metabolites; *Mercurialis perennis*

Postfire succession of oak forests and scrubs of hornbeam on the Vidlič Mountain (southeastern Serbia)

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On the Vidlič Mountain in southeastern Serbia in the summer of 2007. the wildfire occurred, in which burned beech and oak forests, scrubs of hornbeam, dry pastures and rocky ground vegetation. The influence of wildfire on oak forests and hornbeam the first three years after fire was followed. At localities where oak forest burnt completely it is a different physiognomy compared to the situation before the fire. The initial stage of succession is characterised by the presence of annual species to less altered habitats at the medium altitudes. The next stage is with the dominance of perennial plants. The floristic diversity increases with altitude. At the sites where it has happened only partially damaged, the ecosystem recovery is faster and the time to establish the initial state is shorter. Oak forests with hornbeam scrubs are thermophilic type, and the land is not developed and porous. Therefore, the succession is very slow process that takes place for decades.

Keywords: fire, Vidlič, oak forests, scrubs of hornbeam, diversity

Variability of morphological characters of the genus *Mentha* L. in Republic of Macedonia

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Genus *Mentha* L. comprises species with high content of volatile, aromatic, essential oils hence they are medicinally and economically very valuable, so traditionally, chemical content of its essential oils, alcohols extracts, phenols and flavonoids, but also antifungal and antioxidative activities are the main topics of investigations worldwide. On the other hand, genus is taxonomically complicated, highly variable and species hybridize often and easily with each other. In Republic of Macedonia complete critical taxonomic revision of the genus that include morphometric analysis has not been done so far. New taxa of the genus have been recorded during floristic and phytocoenological investigations, and more recently in ethnobotany studies. Additionally, large trouble is lack of volume of the Flora of Republic of Macedonia with family Lamiaceae, due to there are unknown actual number of taxa and unclear distribution and habitat patterns of genera and species. Therefore, during 2013 and 2014 in summer vegetation seasons we investigated few localities in west and central part of the country and

collected plant material. In total, we collected six taxa, predominantly from moist, wet and damp habitats near lakes, rivers or channels. We conducted morphological study on five species *Mentha aquatica*, *M. longifolia*, *M. microphyla*, *M. spicata*, and *M. pulegium*, and on one hybrid *M. x dumetorum*, using characters of vegetative and generative regions of plants. Results of measurements were statistically analyzed in STATISTICA ver. 12 and PAST ver. 3 softwares. Analyzed samples follow emphasized hypervariability of the genus, and we detected and marked characters which contribute the most to that status, but also characters that lead to potential infraspecific differentiation. For clear differentiation of taxa and complete taxonomical-ecological analysis of the genus, it is necessary to increase number of localities and populations from all habitat types that species of the genus inhabit, along with expanding analyzed geographical area and using additional botanical methods.

Keywords: Lamiaceae, mint, traditional morphometry

Morphometric analysis of *Prospero autumnale* (L.) Speta (Hyacinthaceae) complex in Pannonian Basin and Balkan Peninsula

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Beside the two chromosomally stable species of the Mediterranean genus *Prospero* Salisb. (*P. obtusifolium* (Poir.) Speta and *P. hanburyi* (Baker) Speta) there is a karyologically very dynamic and heterogeneous *P. autumnale* complex. For this reason, over the area of distribution (from the atlantic coast and southern parts of England in the west, all over the Mediterranean, including Pannonian Basin in the north and to the Caucasus in the east), have been described numerous species, particularly in the south of the Balkan Peninsula. The existence of these species cannot be completely morphologically supported, because individuals from all over the area are morphologically very similar, with slight differences mainly in quantitative characters (dimension of leaf, plant height, flower size and filament width). In this study, morphological variability of 537 individuals from 19 populations from Panonian Basin and Balkan Peninsula, which have been selected as a priori defined groups, were analyzed. The dissected parts of plant were scanned and measured using Digimizer software 4.5.2. A total of 21 quantitative characters (2 characters of vegetative and 19 characters of generative region) were analyzed and statistically processed using program Statistica ver. 13. The results of multivariate statistical analyses (PCA, DA) confirmed the existence of small differences among populations of this complex, also at the same time results point to the slight separation of the pannonian, western Balkan and south Balkan populations from the populations from middle part of Balkan Peninsula. Characters that mostly contributed to the discrimination among the analyzed populations were style and filament length, plant height and the ratio between lengths of style and ovary.

Keywords: morphological variability, quantitative characters, multivariate statistical analyses

Heavy metal contents in soils and plants of *Viola kopaonikensis* Pančić ex Tomović & Niketić and *V. tricolor* L. from eight ultramafic sites in Serbia

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It is well known that some *Viola* species can accumulate trace elements in their aboveground parts, and can act as hyperaccumulators of Zn, Cd or As. The aim of the present study was to determine heavy metals contents in soils and plants belonging to two *Viola* species (*V. kopaonikensis* and *V. tricolor*) from eight ultramafic localities in Serbia, in order to examine their accumulation potential. Chemical characteristics of the soil samples (pH, % of organic C, P₂O₅, K₂O, Ca, Mg, Fe, Mn, Ni, Zn, Cu, Cr, Co, Cd and Pb) and plant roots and shoots (P₂O₅, K₂O, Ca, Mg, Fe, Mn, Ni, Zn, Cu, Cr, Co, Cd and Pb) were determined. Also, for comparison of two species, bioconcentration and translocation factors were used, as well as Spearman's rank correlation coefficients for concentrations of elements in root and shoot samples. Species *V. kopaonikensis* acted as strong Ni and Cr accumulator, with high concentrations of Ni in the shoots and concentrations of Cr in the aboveground plant tissues in samples from two localities several times higher than expected. Populations of *V. tricolor* showed no potential for trace element accumulation and belong to the group of species tolerant to ultramafic soils. This species can be considered as heavy metals excluder.

Keywords: trace elements, accumulator, excluder

Anatomical characteristics of *Glaucosciadium cordifolium* (Boiss.) Burt et Davis (Apiaceae)

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Glaucosciadium is a monotypic genus including only *Glaucosciadium cordifolium* which distributes in the Mediterranean region. The aim of this study is to determine anatomical properties of this genus for the first time. For this purpose, cross sections of stem, fruit and leaf along with surface sections of leaf are examined in *Glaucosciadium cordifolium*. The leaves are amphistomatic and mesophyll is equifacial. The stem is terete, includes regularly arranged oil ducts with about equal intervals. Along the radial direction, each oil duct is near to the well-developed collenchyma above, and near to a vascular bundle below. The fruit has four vittae on lateral and dorsal faces, but no vittae on commissural face.

Keywords: Anatomy, *Glaucosciadium*, Turkey

Physiological responses and tolerance of black poplar (*Populus nigra* L.) clones to soil water deficit

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Drought is one of the most important factors affecting plant growth, development, survival and productivity. Poplars are among the most drought-sensitive woody plants, and their drought tolerance varies greatly among species, populations and clones due to high genetic diversity. Arid habitats appeared to be more numerous due to effects of global climate changes. Assessment of poplar plants responses to drought plays a major role in breeding programs aimed at production of clones characterized by superior growth and resistance to moderate and/or severe drought. Selection of drought tolerant clones might improve the performance of poplar stands and plantations under arid conditions.

The aim of this study was to quantify some morpho-physiological and biochemical parameters in black poplar clones experiencing different watering regimes.

Five black poplar (*Populus nigra* L.) clones (VII/25, IX/30, I/2, X/32, XI/36) were subjected to different soil water regimes (90-70% of maximum soil water saturation - control, moderate drought followed by recovery of optimal soil water saturation (90-40%) and severe drought (50-40% of soil water saturation)) for 21 days. Plants were grown in a semi controlled environment (greenhouse) by soil culture method. Differences in leaf gas exchange, biochemical and growth parameters were investigated among genotypes and treatments, to reveal plants response to water deficit and recovery from drought. Extended period of drought caused reduction in photosynthetic and transpiration rate in all clones. Considerable increase of these parameters has been recorded after the recovery period. Accumulation of proline in leaves increased along with the intensity of drought. Clones VII/25 and I/2 showed better osmotic adjustments and higher drought tolerance than the other examined clones. Water deficits significantly slowed down apical growth and shoot height growth in all clones except in X/32, while VII/32 and X/32 showed similar number of leaves during all treatments.

The obtained results elucidated the differentiation of studied poplar clones according to their responses and tolerance to soil water deficit.

Keywords: drought, growth, recovery, photosynthetic characteristics

Axenically culturing the bryophytes: establishment and development in *in vitro* condition of two *Ptychostomum* species from Macedonia

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The moss genus *Ptychostomum* comprise over 60 species worldwide, with a primary radiation in the Northern Hemisphere where the highest species diversity can be found in arctic, boreal and alpine

regions. Different ploidy levels and ploidization among species of *Ptychostomum* is often recorded. This can be possibly one of the reasons for ubiquitous behavior in new habitat settlement.

In order to study comparative developmental ecophysiology, we established two species from two section of the genus, namely *Ptychostomum torquescens* and *P. archangelicum*. Plant material for the study was collected in Mavrovo National Park (Macedonia) and species were established under the same conditions. However, it was noted that under same conditions they express different forms of development. The spores of *P. archangelicum* germinate quickly, up to one week after spread on growth media, while the spores of *P. torquescens* needed up to three weeks to start to germinate. Besides, under the same growth condition (18C, and 16/8 h light period) and on the same media type (KNOP), *P. torquescens* remained in the protonemal phase, sometimes developing caulonemas, brachyocytes and tmemas. On the contrary, *P. archangelicum* in the same controlled conditions developed protonema and started bud formation quickly after spore germination. Plantlets grew to fully developed gametophores soon after. Also, it produce secondary protonema upon transfer to new medium.

Here, we give the comparative developmental morphological features for these two species in the frame of the same ecological factors and (prevised) physiological responses.

Keywords: mosses, propagation, growth, comparative ecophysiology

Embryo viability of *Juniperus excelsa* of Prespa area

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Juniper seeds are generally dormant while many of them are empty or contain a non-viable embryo, causing serious difficulties in the species' regeneration. The present research, which was an action of the Life "JunEx - Restoration and Conservation of the Priority Habitat Type *9562 Grecian Juniper Woods in Prespa National Park, Greece", examined the germination and viability of *Juniperus excelsa* seeds. Both mature and immature seeds were collected from the Prespa area. A significant number of seeds were cut manually. The majority of them were empty. The rest of the seeds were divided to mature and immature and the embryos were subjected to Topographical Tetrazolium Test after the end of which embryos' classification as viable and nonviable were evaluated. The percentage of empty seeds was very high ranging from 83% in immature seeds to 90% to mature ones. Only ≈2% viable embryos in mature seeds and ≈5% in immature seeds were recorded in the total seeds lot. From the results we may conclude that the enormous percentage of empty seeds and mainly the extremely small percentage of viable embryos put additional obstacles to the germination of a species well known for its difficult regeneration.

Keywords: viable embryos, empty seed percentage, TTZ test

Production of phenolic compounds and antioxidant activity of *Hypericum dimonieii* Vel. wild-growing plants collected in Macedonia

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The genus *Hypericum* belongs to Hypericaceae, a family composed by species with important medicinal properties. In Republic of Macedonia, the genus *Hypericum* includes 15 species, 3 subspecies, 10 varieties and one endemic species *H. dimonieii*. This study reports for the first time the production of phenolic compounds and antioxidant activity in different parts of *H. dimonieii* Vel. collected from Republic of Macedonia. The methanolic extracts of the aerial parts of the plant, stem, leaf and flower were used for determination of total phenolic and flavonoid contents using Folin-Ciocalteu's and aluminium chloride methods, respectively, while antioxidant activity was assayed using DPPH (2,2-diphenyl-1-picrylhydrazyl) radical scavenging capacity method. The highest content of total phenolics was found in leaf extracts (71.35 mg·g⁻¹ DW), followed by aerial parts of the plant and flowers (about 40 mg·g⁻¹ DW). The lowest content of total phenolics was noticed in stem extracts (32.31 mg·g⁻¹ DW). Also, the highest content of total flavonoids was found in leaf extracts (23.98 mg·g⁻¹ DW). The total flavonoid contents in other tested extracts were ranged from 14 to 17 mg·g⁻¹ DW. The highest antioxidant activity was found in leaves and the aerial plant parts (about 180 µM·g⁻¹ DW), while flowers and stems had low DPPH activity (below 20 µM·g⁻¹ DW). The correlation analyses showed that flavonoids are dominant phenolic compounds responsible for antioxidant activity of tested plant extracts. This study provides some useful insight into the phenolic compounds production and antioxidant potency of different parts of *H. dimonieii* and may justify further investigation of other beneficial biological properties.

Keywords: Antioxidant activity, Flavonoids, *Hypericum dimonieii* Vel., Phenolic compounds.

Production of phenolic compounds and antioxidant activity of selected wild mushrooms from Tricholomataceae family, collected in Macedonia

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Macrofungi have long been valued as nutritional food and an important part of the traditional medicine by different cultures worldwide. They contain a variety of secondary metabolites which have been shown to act as excellent antioxidants. Phenolic compounds are widely distributed secondary metabolites in mushrooms, which are correlated with their antioxidant activity. Considering the growing

interest for the natural sources of antioxidants and the unexplored potential of Macedonian wild fungi, the aim of this study was to investigate the production of phenolic compounds and antioxidant activity of three wild species belonging to two genera of Tricholomataceae family: *Collybia peronata*, *Tricholoma aurantium* and *T. stans*. Antioxidant properties of the selected fungi were assayed using DPPH (2,2-diphenyl-1-picrylhydrazyl) radical scavenging capacity method. Total phenolic content in the methanolic extract of the mature fruiting bodies was determined by Folin-Ciocalteu's colorimetric method, while for the determination of the flavonoid content was used aluminium chloride colorimetric method.

The highest content of total phenolic compounds was found in *Tricholoma stans* (19.43±0.70 mg GAE/g DW), followed by *T. aurantium* and *Collybia peronata* (15.93±0.58 and 10.60±0.26 mg GAE/g DW, respectively). The highest flavonoid content was also determined for the extract of *T. stans* (3.47±0.18 mg QE/g). Among the tested samples, *T. aurantium* fruiting body extract was the strongest DPPH scavenger with the lowest EC₅₀ value (0.30), followed by the extracts of *T. stans* and *C. peronata* (0.32 and 0.61, respectively).

This study demonstrated that the analysed wild macrofungi (*Collybia peronata*, *Tricholoma aurantium* and *T. stans*) have the ability to accumulate phenolics as bioactive secondary metabolites and possess antioxidant activity in *in vitro* systems.

Keywords: Collybia; Tricholoma; Antioxidant; Phenolics; Flavonoids

Molecular determination of fungal strains of *Metarhizium* species isolated in different regions in Switzerland

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The use of entomopathogenic fungi for the biological control of cockchafer grubs dates several decades. Only very few strains of the fungus *Beauveria brognartii* and *Metarhizium* have been commercially used for this purpose. Our results of several studies show the dominant presence of species of the genus *Metarhizium*. In this study isolates of *M.anisopliae*, *M.brunneum*, *M.robetsii* and *M.guizhouense* have been identified. Using elongation factor α (EF α) as PCR primer, different strains of each of the species could be distinguished. *M.brunneum* was present in three isolates: KVL 13-02, KVL 13-03 and KVL 13-06. *M.robetsii* was presented in two different strains: LPSC_963 and ESALQ_1620.

The other species were presented in small numbers which could not be interpreted statistically and have to be repeated with larger amounts of probes.

Visually the different groupings with strains of *Metarhizium* were showed by dendrograms showing the statistical distances (bootstrap) between them.

Keywords: entomopathogenic fungi, PCR, fungal strains, dendrogram.

Which cranial view (dorsal, ventral, lateral or occipital) best reflects phylogenetic relationships among five European grayling (*Thymallus thymallus*) populations?

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The European grayling, *Thymallus thymallus* (Linnaeus, 1758) is salmonid fish species with complex life history depicted mostly by recent genetic research. As a result, at least five major phylogenetic lineages have been described and fine genetic structure in each of the lineages has been discovered. Herein, we analyzed cranial morphology of *T. thymallus* from three different basins that correspond to three previously described phylogenetic lineages. Landmark-based geometric morphometric methods were applied to four cranial views (dorsal, ventral, lateral and occipital). We used specimens from five European grayling populations (Adriatic Sea – Soča River, Black Sea – Sava Bohinjka and Una Rivers, Caspian Sea – Bugurla and Kana Rivers). Cranial size analyses revealed significant size differences for all pairwise comparisons (dorsal cranium: $P < 0.05$; ventral cranium: $P < 0.01$; lateral cranium: $P < 0.05$; occipital cranium: $P < 0.05$). When comparing centroid sizes (CS) for all four views, Soča population had the largest mean values, while Bugurla population had the smallest values of centroid size means. Significant interaction between log CS and population was revealed only for ventral cranium ($\lambda_{\text{Wilks}} = 0.1454$, $F_{92,232,07} = 1.58$, $P = 0.0031$). Therefore, Canonical variate analysis (CVA) of inter-population ventral cranial shape differences was conducted without correction for the allometry. For the other three cranial perspectives, CVA of non-allometric shape variation showed best separation of studied populations at the level of occipital cranium. Namely, populations from the same basin were grouped more tightly, while Soča population was separated from the other four populations. Specimens from Caspian Sea basin showed overall flattening of the cranium in occipital view. In individuals from Soča population dorsal base of supraoccipital crest was moved more dorsally, resulting in higher skull with pronounced supraoccipital crest. Most of the landmarks digitized on occipital cranium were located on neurocranial skeletal elements that are generally considered more conservative and thus gave best reflection of phylogenetic relationships.

Keywords: Salmonidae, geometric morphometrics, allometry, morphology, shape, skull, size

Morphological variation of European grayling (*Thymallus thymallus*) from three different basins (Adriatic Sea, Black Sea and Caspian Sea) based on external body morphology

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The European grayling, *Thymallus thymallus* (Linnaeus, 1758) is salmonid fish species naturally inhabiting most of European continent. In the past two decades majority of research aimed European grayling genetic structure and description of its phylogenetic lineages. In contrast, not many morphological studies of this species have been published so far. We analyzed external body morphology of *T. thymallus* from three different basins that correspond to three previously described phylogenetic lineages. Landmark-based geometric morphometric methods were applied to specimens of five European grayling populations (Adriatic Sea – Soča River, Black Sea – Sava Bohinjka and Una Rivers, Caspian Sea – Bugurla and Kana Rivers). Body size analyses disclosed statistically significant differences for all pairwise comparisons, except for those between Sava Bohinjka and Una ($P=0.0505$) and between Bugurla and Kana populations ($P=0.1016$). When comparing centroid size (CS), Soča population had the largest mean value ($CS=40.11$), while Bugurla population had the smallest one ($CS=18.67$). Using multivariate regression of shape variables onto log CS we found that allometric effect was significant ($P<0.0001$) and accounted for 6.37% of overall shape variation. Canonical variate analysis (CVA) of non-allometric shape variation showed clear separation of all populations. Specimens from Bugurla and Kana populations in comparison to those from the other three populations (separated along CV1) are characterized by heads with smaller opercle in relation to subopercle and trunks with longer dorsal and adipose fins and larger distance between anterior bases of ventral and anal fins. Individuals from Soča population compared to those from Una population (separated along CV2) have more robust heads and trunks with longer dorsal fins. These results match to genetic differences already observed for populations studied herein. Morphologically most diverged population was the one from Soča River (Adriatic basin) which is in accordance with its large genetic distinctiveness.

Keywords: Salmonidae, geometric morphometrics, allometry, shape, size

Morphological Differences of Brown Trout (*Salmo trutta* L.) Populations from Serbia and Bosnia

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Using geometric morphometrics, 45 individuals of brown trout (*Salmo trutta* L.) from four localities in Serbia and Bosnia and Hercegovina were analyzed, in order to estimate the differences between populations. Samples were collected from one location in Bosnia and Hercegovina: Una and its tributaries, and three locations in Serbia: Đerdap National Park, River Rasina near Kruševac in Central Serbia and River Radovanska near Zaječar in Eastern Serbia. Size and shape of analyzed individuals was described by 14 landmarks using programs tpsDig2 1.38, tpsUtil 2.10 and MorphoJ 1.02i. Analysis of variance (ANOVA) showed significant differences of centroid size between populations ($F = 15.13$; $df = 3$; $p < 0.00001$). The same significant result was also revealed by multivariate analysis of variance (MANOVA) using Procrustes coordinates (Wilks' Lambda = 0.022280; $F = 1.7$; $p = 0.000000$). Canonical Variate analysis (CVA) allocated Radovanska and Una from Rasina and Đerdap populations according to first axis that holds the highest percent of variability (80.6%). Second axis that holds 11.7% and third axis that holds 7.7% of variability separated River Una from River Radovanska population and River Rasina from Đerdap population, respectively. Nevertheless, Cluster analysis based on Procrustes distances showed similarities between Rivers Radovanska and Una as well as between Đerdap and River Rasina populations, which was in accordance to the results of the first CVA axis. Such clustering could be explained by ecological similarities of analyzed localities that influenced trout morphological characteristics.

Length-weight relations and conditional state of common roach (*Rutilus rutilus*), common bream (*Abramis brama*) and their natural hybrid from the accumulation Modrac

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In the accumulation Modrac there has been, for the first time, registered presence of natural hybrid of roach and bream. During the research there has been collected 1103 individuals of different fish species, among which roach was the largest group with 345 individuals (31,25%). Bream was represented with 216 individuals (19,56%), and hybrid with 96 individuals (8,7%). Total ichthyomass of the roach has been established, and it was 9694,57 g (13,8%) of total ichthyomass. Ichthyomass of the bream was 27901,24 g (39,72%), while ichthyomass of the hybrid was 9071,15 g (12,91 %) of total ichthyomass. Total body length of the roach had its limits of variations between 5,00 – 22,50 cm ($\bar{x} = 11,85$), standard body length between 4,10 – 18,50 ($\bar{x} = 9,64$), and ichthyomass between 0,70 – 222,70 g, ($\bar{x} = 27,77$). Total

body length of the bream varied between 8,20 – 40,00 cm ($\bar{x} = 26,2$), and standard length between 6,20 – 32,50 cm ($\bar{x} = 17,78$). Value of individual ichtyomass varied between 5,60 – 625,00 g ($\bar{x} = 12,16$). Total body length of the hybrid was between 9,90 and 23,00 cm ($\bar{x} = 19,51$), with standard lengths between 7,40 – 18,40 cm ($\bar{x} = 15,57$). Value of individual hybrid ichtyomass varied between 8,70 – 124,60 g ($\bar{x} = 84,7$ g). Length-weight relations were calculated using this formula $r = \frac{\sum[(x-\bar{x}) \cdot (y-\bar{y})]}{N \times S_x \times S_y}$. Data of length-weight relations had served in calculating the condition factor (CF), which was calculated using this formula $CF = W \times L^{-3} \times 100$.

Keywords: roach, bream, hybrid, length-weight relations, condition factor

Age Structure of the Endemic Fish *Phoxinellus pseudalepidotus* (Cyprinidae) from Mostarsko Blato (Neretva River Basin, Bosnia and Herzegovina)

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This study provides age structure information for *Phoxinellus pseudalepidotus*, a freshwater fish endemic to the Neretva River basin, which is classified by the IUCN as vulnerable. Fish sampling was carried out in 2009 by gill nets and „krtoľ“, a traditional hunting tool in the area of Mostarsko Blato (Neretva River Basin, Bosnia and Herzegovina). In order to analyze age structure of the population, 70 units of various age categories were taken. Length range of all analyzed units was from 2,7 to 11,5 cm. By reading otoliths, a total of four age classes were established: 0+, 1+, 2+ and 3+. The largest part of the sample consisted of 2-year-old units (40%). There were 28,6% of 1-year-old units, 27,1% of 3-year-old units, and 4,3% of 4-year-old units. The mean estimated total length (TL) of 1-year-old *P. pseudalepidotus* was 4 cm; 2-year-old, 6,7 cm; 3-year-old, 9,15 cm; 4-year-old, 11,05 cm. Similar data on the oldest age were found for the species *Pelagius epiroticus*, an endemic species widespread in Lake Pamvotis in Greece. This species grows to about 5-6 cm of total body length in the first year of life, and the maximum registered age was 2 to 3 years. On top of that, for the species *Anaocypris hispanica*, widespread in the Guadiana River Basin in Spain and Portugal, the oldest registered age was 3 years. For the species *Telestes montenegrinus* from the Morača River in Montenegro seven age classes can be distinguished. By determining the age of *Telestes ukliva*, from the Cetina River, eight age classes were detected, from 0+ to 7+. The oldest registered age class in *Telestes souffia* from the Drina River was 7+. It can be concluded from the above that *P. pseudalepidotus* is a short-living species which leaves four years at the longest. The conducted study provides an insight into age structure of *P. pseudalepidotus* population, which represents new and original contribution to knowledge about this endemic fish.

Keywords: *Phoxinellus pseudalepidotus*, age structure, Mostarsko Blato

Morphological characters of lacustrine and riverine populations of *Aulopyge huegeli*

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Dalmatian barbelgudgeon, *Aulopyge huegeli*, the only representative of its genus, is endangered, strictly protected freshwater fish species endemic to the Dinaric karst. It lives only in few rivers, lakes and sinkholes in Croatia and Bosnia and Herzegovina (Cetina, Krka, Čikola, Buško Blato and Livanjsko, Duvanjsko and Glamočkopolje). Knowledge about this unique fish species is scarce – there is a little data about its biology, ecology and phylogeny, what makes this species vulnerable to different types of threats. It is mostly threatened by dam construction, water extraction and pollution, habitat loss and degradation, changes in water regime and, especially, by excessive exploitation. In last few decades a substantial decline in population of *Aulopyge* is recorded. It is known that there can be certain differences in morphology and ecology among riverine and lacustrine populations of fish species. Therefore, the aim of this study is to verify if this is the case with *A. huegeli*. Specimens were collected by electrofishing and gill nets from four different locations representing lakes (Visovac Lake on River Krka, Toraklake on River Čikola) and rivers (River Čikola, channel Ždralovac). Twenty four morphometric characters were measured, standardized and analysed with statistical methods (descriptive statistics, ANOVA and PCA). In addition to morphometry, meristic characters and external appearance were also included. Meristic analyses included fin rays from all fins (dorsal, anal, caudal, pectoral and ventral fin) and external appearance description of colouration and body shape of specimens from different locations. The preliminary results show differences between males and females as well as between populations of *A. huegeli* from different habitats – lacustrine and riverine. The latter confirms earlier stated hypothesis of species' adaptation to different habitat types.

Keywords: Dalmatian barbelgudgeon, morphology, Croatia, Bosnia and Herzegovina

Winter Diet of Otters in the North Western Serbia, a New Parameters and Methodological Perspectives

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Diet of otters reflects their habitat requirements, habitat quality and their impact on prey, which generates a need for a reliable method of quantifying prey categories in their food.

The aims of this study were a) to determine the level of mutual compatibility of results obtained per one sample, by using two most common methods of quantification of broad taxonomic categories (fish, frogs, reptiles, insects, crustaceans, mammals): the relative frequency of occurrence (RFO) and

the relative percentage of weight (RWP) and b) the identification and analysis of the reliability of fish species determination and quantification depending on the structures on which their classification is based on.

The material analyzed consisted of 192 otter faeces samples. The following analysis was made: a) pooled samples from two methods were shown as two vectors with the same number of components (broader taxonomic groups) where the angle between them was a measure of their similarities b) an index of sums of scales and vertebrae presence was designed in relation to bone elements, as an assessment of species determination.

Compared methods with untransformed variables were harmonized until the exclusion of fish and an additional execution of a transformation. The maximum index value was present in the family Esocidae, while a minimum frequency of scales and vertebrae was noted in the Cyprinidae family.

The two methods can be replaced by each other as long as the fish is the dominant prey. Since the vertebrae and scales do not make it possible to determine the number of individuals, there can be an overestimation of the presence of small prey, and these structures are extendedly eliminated in a few faeces samples, leaving the frequency of species with high index values in question. It would, therefore, be potentially useful to specify subtle differences between the vertebrae along the spinal column for the quantification of prey individuals.

Keywords: fecal analysis, method comparison

Increase of planktonic Tunicata (Appendicularia, Thaliacea) biodiversity in the Adriatic Sea: A possible relationship with hydroclimatic changes in the Mediterranean Sea

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We investigated potential connections over the past two decades between mesoscale circulation regimes in the Ionian Sea and newly-observed planktonic tunicate species and concurrent rise in sea temperature in the Adriatic Sea. Analyses of plankton samples from 1993 to 2015 in the southern Adriatic revealed marked changes in the planktonic tunicate community. Eleven appendicularian and three thaliacean species were recorded for the first time in the Adriatic. We found that incoming of these species are in connection with circulation regimes in the Northern Ionian Gyre (NIG). The occurrence of Atlantic/Western Mediterranean species coincided with anti-cyclonic circulation in the NIG, owing to the advection of Modified Atlantic Water into the Adriatic, while the presence of Lessepsian species coincided with the cyclonic pattern, which governs the entry of Eastern Mediterranean waters. The impact has been that some newcomers now make a larger contribution to the zooplankton community in the southern Adriatic and, in certain cases, have replaced native species. The synergistic effects of these processes, together with warmer Mediterranean waters, raise concerns over dramatic changes in the marine biodiversity of the Adriatic.

Keywords: Ionian Sea, planktonic tunicate species, sea temperature, Adriatic Sea, zooplankton community

Length-weight relationship and condition factor of *Barbus balcanicus* in fragmented and non-fragmented habitats in central Serbia

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The aim of this study was to describe the differences in Length–weight relationship (LWR) and Fulton's condition factor (K) of large spotted barbell (*Barbus balcanicus* Kotlík, Tsigenopoulos, Ráb & Berrebi, 2002) caused by stream damming. A total of 887 specimens (4.2–19.4 cm total length) used in this study was collected on small rivers in the central part of Serbia in 2014. The value of regression coefficient (b) obtained for LWR varied from 2.63–3.67±0.09. In non-fragmented habitats suggests positive allometry growth. The result showed a negative allometric growth in population on localities where streams are impounded. The regression coefficient values for specimens in non-fragmented habitats are higher than 3, and that suggest positive allometric growth. The condition factor calculated for *B. balcanicus* varied from 0.84–1.14±0.27 and indicated that the specimens were moderately healthy. This study has contributed to the knowledge of impact stream damming on populations of *B. balcanicus* in this area, which could assist scientists in carrying out future ecological studies in line with the strategies of conservation, restoration and management.

Keywords: large spotted barbell

Examining heavy metal accumulation potential of *Viscaria vulgaris* Bernh. (Caryophyllaceae) from four ultramafic localities in Serbia

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Many representatives of the family Caryophyllaceae withstand the challenges of harsh conditions in ultramafic habitats. Our aim was to examine trace element profiles of *Viscaria vulgaris* from four ultramafic localities in Serbia (Mt. Maljen, Mt. Zlatibor, Mt. Stolovi and Mt. Vujan) and its possible accumulation potential. Chemical characteristics of the soil samples (pH, % of organic C, and concentrations of P₂O₅, K₂O, Ca, Mg, Fe, Mn, Ni, Zn, Cu, Cr, Co, Cd and Pb) and plant samples (concentrations of P₂O₅, K₂O, Ca, Mg, Fe, Mn, Ni, Zn, Cu, Cr, Co, Cd and Pb), accumulation and translocation factors were determined. Correlations between concentrations of analysed elements in root and shoot samples were performed using non-parametric Spearman rank-order correlation. According to Kruskal–Wallis ANOVA all samples differed significantly among localities in trace element concentrations. Only Ni and Cd concentrations in root samples did not differ significantly. Although concentrations of Ni in all root samples exceeded 100 mg kg⁻¹, the most of Ni was not transported into the shoots and therefore *V. vulgaris* can only be considered as Ni excluder. Concentrations of Cd in all plant samples were higher than available concentrations in soils, but they did not fall in range of values

considered excessive or toxic for plants. No potential for heavy metal accumulation in *V. vulgaris* was determined in this study.

Keywords: excluders, trace elements, ultramafics

Genetic characterization of European mudminnow (*Umbra krameri*) population from Special Nature Reserve „Kraljevac“, Serbia

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U. krameri exists at three locations in Serbia, one at Bakreni Batar in the Sava River system, and two at Lugomir and Kraljevac in the Danube River drainage. Population from Kraljevac (close to village Deliblato) is recorded last, in December 2014. From mentioned populations, only population from Kraljevac has not been genetically analysed. Genetic characterization of Kraljevac population will help in future protection and conservation of this critically endangered species in Serbia. Genetic structure of Kraljevac population was studied by analysing the mitochondrial cytochrome b gene (1085 bp) and the seven microsatellite loci. Ten individuals were analysed and three cytochrome b haplotypes detected (Da1, Da3 and Sa1), suggesting Danube and Sava phylogeographic lineages. The most frequent haplotype was Da1 (50%), followed by Da3 (30%) and Sa1 (20%). All three haplotypes are already known, from the Upper and Middle Danube (Da1), from the Middle and Lower Danube (Da3) and from the Sava River system (Sa1). Microsatellite analysis was performed on 24 individuals, and this population is characterized by the high parameters of genetic diversity ($H_e=0.762$, $H_o=0.798$, $A=9.29$ and $Ar=8.63$) in comparison to populations from Bakreni Batar ($H_e=0.766$, $H_o=0.779$, $A=8.14$ and $Ar=8.03$) and Lugomir especially ($H_e=0.634$, $H_o=0.639$, $A=5.86$ and $Ar=5.86$). The pair-wise F_{st} comparison test reveals a statistically significant difference between all three Serbian populations ($P<0.01$). Moreover, the difference between Kraljevac and Bakreni Batar populations is about 4.5 times lower, in relation to Kraljevac and Lugomir. Higher relatedness among these two geographically closer populations was also evident from the Das distances and Factorial Corresponding Analysis. To conclude, Kraljevac population represents a unique and natural genetic mix among Danube and Sava mudminnow populations, and as such deserves a special conservation status.

Keywords: *Umbra krameri*; mitochondrial DNA, microsatellites; conservation, Danube River drainage

Nickel hiperaccumulation by the species of *Alyssum murale* group from Macedonia

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Alyssum murale and related species belong to the section *Odontarrhena*. There is a big debate in the scientific literature whether these taxa deserve specific rank or they represent the varieties in the frame of the broadly understood *A. murale* species. However, the section was recently raised to the level of genus and many of the described taxa received or were accepted at the specific rank by the Euro+Med PlantBase. The species described by professor Micevski from Macedonia (*Alyssum serpentinum*, *A. kavadarcensis*, *A. debarensis*, *A. gevgelicensis* and *A. skopjensis*) are among them under the genus name *Odontarrhena*.

The aim of the presented work was to identify the potential of the species of *A. murale* group for Ni hiperaccumulation. Beside the above mentioned species, other species of the new genus *Odontarrhena* distributed in the Republic of Macedonia: *O. chalcidica* (= *A. subvirescens*) and *O. muralis* (= *A. murale* and *A. pichleri*) were analyzed too. Additional target of the research was to reveal possible differences regarding Ni accumulation among different investigated species. Several other species: *Alyssum striberni* and *A. strigosum* as well as *Odontarrhena corimboideia* (= *A. corymbosoides* and *A. vranjanum*) were analyzed for the meter of comparison.

The results showed high level of hiperaccumulation of Ni by all investigated *Odontarrhena* species, except *O. corymbosoides*. The average values of Ni content range from 320 mg·kg⁻¹ (*A. pichleri*) to 5329 mg·kg⁻¹ (*A. serpentinum*). There is a significant correlation between Ni content in the leaves of all species and in the respective soils.

Keywords: *Alyssum murale*, Ni, hiperaccumulation

Assessing the watershed habitat evaluation and stream integrity protocol (WHEBIP) applicability in assessing stream integrity in river Bregalnica watershed

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This paper presents the results of the rapid assessment of ecological integrity of a total of 1421 stream segments from more than 250 streams in river Bregalnica watershed with emphasis on river

Bregalnica as the biggest and most important watercourse in Eastern Macedonia.

The ecological integrity of streams in Bregalnica watershed is calculated by estimating the land use impact on adjacent riparian habitats, and the impact that multiscale environmental properties have on sub-basin features. The results have principally been derived from remote sensing data and set up in a model build up on Watershed Habitat Evaluation and Stream Integrity Protocol (WHEBIP). WHEBIP effectiveness in predicting ecological integrity of streams has been assessed by statistical analysis of correlation derived upon available data on macroinvertebrate biotic indices and physical-chemical parameters on 35 localities throughout the basin.

Statistical analyses showed a strong correlation between WHEBIP integrity scores and macroinvertebrate biotic indices thus confirming the capacity of WHEBIP to predict stream site-specific features with great accuracy in case of Bregalnica. The results obtained in this study contribute towards improvement of the WHEBIP protocol and in general promotes applicability of rapid stream integrity assessment tools in setting priorities for integrated management and conservation plan for watersheds and streams.

Keywords: Bregalnica, basin, sub-basin, stream assessment, macroinvertebrate, WHEBIP

Genotoxicity tests and their contributions in ecotoxicological research

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The cytokinesis-block micronucleus cytome assay (CBMN) is suitable and standardized test in genotoxicology used for assessment of genotoxicological effects of different chemical or physical agents and it is the most commonly used method for measuring DNA damage in human lymphocytes. The micronucleus assay, like basic genotoxic test in molecular epidemiology and cytogenetics, is applied as diagnostic tool and procedure for measuring micronucleus (MN) frequency in peripheral blood lymphocytes (PBL). The aim of this study was to evaluate the genotoxicity of ionizing radiation using the CBMN assay and to determine the human health risk. The study population included 20 healthy workers exposed to ionizing radiation (radiologist, technicians and nurses) and 20 individuals healthy people not exposed on radiation. 12 individual samples (60 %) showed an increase in the MN frequency while in the control group, the increased frequency of MN_i was found in 1 of the blood samples (5%). These results suggest that chromosomal instability is in the correlation with micronucleus frequencies and genetic load in healthy workers. This study has a practical importance because it indicates the necessity of introducing a permanent genotoxicological and other monitoring on the vulnerable category of workers (in this study medical workers).

Keywords: genotoxicology, DNA damage, human lymphocytes, ionizing radiation

Development of Naringenin-Modified Glassy Carbon Electrode for Electrochemical Determination of Copper (II) Ions: Application in Beyşehir Lake Water Sample

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Trace metals commonly exist as environmental pollutants. These pollutants can easily be taken up by mankind, animals, plants and waters in the environment. These metals enter human bodies through water and food, thus copper determination in these samples is very important. Electrochemical modification of glassy carbon (GC) electrode by naringenin (NG), a flavonoid derivative, and applicability of electrode modified in this way for determination of copper (II) (Cu (II)) ions is reported in this study. Surface modification experiments were performed in phosphate buffer solution (PBS), pH 7, in the 0.0 mV and +1400 mV potential ranges with a sweep rate of 100 mV s⁻¹ and 30 cycles. The surface modification of GC electrode was performed with NG using cyclic voltammetry (CV), whereas the characterization of this sensor electrode was performed using CV and electrochemical impedance spectroscopy (EIS). The sensitivity of GC electrode modified in described way towards Cu (II) ions was investigated in Britton-Robinson (BR) buffer solution, pH 5, by differential pulse voltammetry (DPV). For the calibration curve, a series of standard Cu (II) solution from 1.0x10⁻¹² M to 1.0x10⁻⁶ M was prepared. Detection limit was obtained as lower as 1.0x10⁻¹² M. The proposed method was simple, rapid, low cost and sensitive for the determination of Cu (II) ions.

The proposed method was successfully applied for the determination of Cu (II) ions in Beyşehir lake water sample in Konya, Turkey without any pretreatment. After the grafting of the Cu (II) ions in lake water sample to the NG/GC electrode surface, peak current value was measured by DPV. And then, obtained peak current value was used to detection of Cu (II) ions concentration in lake water sample by interpolating using the calibration curve. The concentration of Cu (II) ions in the real sample is found to be 0.327 µM mL⁻¹ from calibration curve.

Keywords: Flavonoid, Cu (II) ions, Surface Modification, Surface Characterization, Differential Pulse Voltammetry

Aminophenol Modified Glassy Carbon Sensor Electrode: Determination of Phenol in Soil Sample as an Environmental Pollution

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Electrochemical methods are based on the direct oxidation or reduction of substrate onto an electrode surface. Electrode reactions are very suitable for analytical applications due to their requirements of high potential. Moreover, these surfaces can be modified by a reductive substrate for analytical applications. In this study, a simple and sensitive method for the electrochemical determination of phenol

in soil sample using disposable screen-printed glassy carbon (GC) electrode surface modified with an electrografted aminophenol film, via the electrochemical reduction of its prepared aminophenol diazonium salt in non-aqueous media, is presented. 1 mM of aminophenol diazonium salt (APDAS) in 100 mM tetrabutylammonium tetrafluoroborate (NBu_4BF_4) was used for modification of GC electrode surface in this study. The surface modification of GC electrode was performed with aminophenol (AP) using cyclic voltammetry (CV), whereas the characterization of this sensor electrode was performed using CV and electrochemical impedance spectroscopy (EIS). The modification process was carried out in non-aqueous media, whereas phenol determination was carried out in aqueous media. As the final of this study, a reaction mechanism was suggested for the binding of phenol to aminophenol modified GC electrode surface. 0.1316 mg L⁻¹ phenol was determined in soil sample by using AP modified GC electrode.

We tried to show that this recent powerful technique can be applicable to the determination of phenol at trace level. Voltammetric techniques are advantageous to the others because they are inexpensive and reliable. Besides all colored and turbid solutions can be easily analyzed using voltammetric techniques. For this reason, we tried to develop a specific sensor electrode for the determination of phenol in aqueous media by modifying GC surface using APDAS in non-aqueous media. This method can be used in the determination of not only for phenol but also for phenol derivatives.

Keywords: 2,4-diaminophenol, Aminophenol Diazonium Salt, Cyclic Voltammetry, Glassy Carbon Sensor Electrode.

Procaine Modified Carbon Paste Electrode Surface by Cyclic Voltammetry: Quantitative Determination of Phenol in Natural Decayed Leaves

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In this research, the amount of phenol in decayed leaves was quantified in aqueous medium by using procaine hydrochloride modified carbon paste electrode (CPE). Working electrode was prepared by using homogeneous mixture of graphite powder and mineral oil (80:20%). The mixture was pressed on the electrode surface thus a glassy form was obtained. The electrode surface was modified with procaine hydrochloride in 0.1 M tetrabutylammonium tetrafluoroborate using 100 mV s⁻¹ scanning rate between -1.7 V and -0.5 V with 50 cycles. For the calibration curve, a series of standard phenol solution between 1.0×10⁻³ M and 1.0×10⁻⁸ M was prepared using Britton–Robinson buffer solution at pH 12. The prepared standard solutions were used for the chemical binding of phenol to the modified electrode surface between 0.0 V and +0.9 V potential range using 100 mV s⁻¹ scanning rate with 10 cycles. Calibration curve was obtained by plotting concentrations versus peak currents of standard phenol solutions. By using this calibration method, the amount of phenol was determined as 8.33×10⁻⁵ M in natural decayed leaves in BR buffer solution at pH 12. Detection limit was obtained as low as 1.0×10⁻⁸ M. We tried to develop a specific sensor electrode for the determination of phenol by modifying CPE surface using procaine in non-aqueous solution. This study was successfully applied to the real samples for phenol determination. The suggested method can also be applied to drinking water, waste water, soil and food samples in which preconcentration might be needed in some cases although we applied to the decayed leaves. This method can be used in the determination of not only for phenol but also for

phenol derivatives. Lastly, we developed an electrochemical sensor for this quantification. By using this developed sensor electrode one can easily quantitatively determine phenol at very low concentrations.

Keywords: Procaine hydrochloride, Cyclic voltammetry, Carbon paste electrode, Surface modification, Surface characterization.

Investigation of Electrochemical Behaviors of 3,3'-Diaminobenzidine Modified Glassy Carbon Sensor Electrode: Determination of Quantitative Phenol in Tap Water Samples

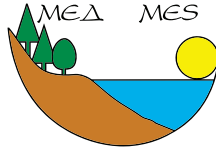
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We tried to show that this recent powerfully technique can be applicable for the determination of phenol at low concentrations. Voltammetric techniques are advantageous to the others because they are inexpensive and reliable. For this reason, we tried to develop a specific sensor electrode for the determination of phenol by modifying glassy carbon (GC) electrode surface using 3,3'-Diaminobenzidine (DAB) in non-aqueous media. This study was successfully applied to the natural samples for phenol determination. In study, the amount of phenol in Meram, Karatay and Selçuklu regions in Konya tap water samples was quantified in aqueous medium by using DAB modified GC electrode. The electrode surface was modified with DAB in 0.1 M tetrabutylammonium tetrafluoroborate (NBu_4BF_4) using 100 mV s^{-1} scanning rate between -0.5 V and $+1.5 \text{ V}$ with 10 cycles. For the calibration curve, a series of standard phenol solution between $1.0 \times 10^{-12} \text{ M}$ and $1.0 \times 10^{-3} \text{ M}$ was prepared using pH 12. The prepared standard solutions were used for the chemical binding of phenol to the modified electrode surface between 0.0 V and $+1.1 \text{ V}$ potential range using 100 mV s^{-1} scanning rate with 10 cycles. By using this calibration method, the amount of phenol was determined as $1.23 \times 10^{-10} \text{ M}$, $3.03 \times 10^{-8} \text{ M}$ and $1.99 \times 10^{-9} \text{ M}$ in natural tap water samples in Britton–Robinson (BR) buffer solution at pH 12. Detection limit was obtained as low as $1.0 \times 10^{-10} \text{ M}$. The presence of DAB, phenol at the GC surface and phenol of DAB modified GC electrode surface was characterized by cyclic voltammetry (CV) and electrochemical impedance spectroscopy (EIS).

This method can be used in the determination of not only for phenol but also for phenol derivatives. Lastly, we developed an electrochemical sensor for this quantification. By using this developed sensor electrode one can easily quantitatively determine phenol at very low concentrations.

Keywords: 3,3'-Diaminobenzidine, chemical sensor electrode, quantitative determination, phenol, tap water.



Section 3
**BIODIVERSITY AND CONSERVATION
BIOLOGY**



Status of roe deer, chamois and brown hare in the potential Balkan lynx recovery areas

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The Balkan lynx population has drastically declined in the past century and the latest assessments show that they have a Critically Endangered status according to IUCN Red List standards. Down-listing its threatened status through population recovery is an urgent objective if we are to preserve the Balkan lynx. To plan the recovery of the Balkan lynx, we created a spatially explicit model which helped us to define the potential recovery area and the recovery routes (corridors). However, the distribution and survival of the Balkan lynx, like most other carnivores, is highly dependent on prey. Recent radio-telemetry research of the Balkan lynx has revealed that roe deer is its main prey, while chamois and hares constitute smaller portion of its diet. Thus, the recovery of the Balkan lynx in the unoccupied areas will mainly depend on the availability of its main prey. Lack of information and research on the distribution and density of the three main prey species (roe deer, chamois and hares) represents a hindrance in predicting the potential success of Balkan lynx recovery. The only available standardised data for Balkan lynx prey is the results from the Baseline survey conducted in the known Balkan lynx distribution area (Macedonia, Albania, Kosovo and Montenegro). This survey provides data on the presence, relative abundance and trends of the three prey species in the assumed Balkan lynx potential distribution area. By comparing the two datasets: the recovery model of the Balkan lynx and the Baseline survey prey data (presence, relative abundance and trend), we obtained spatial information on the areas where the lynx recovery may be successful or hindered by lack of prey availability. The identified areas are potential pilot areas for more detailed research of the prey species and improvement of the status and management of the main Balkan lynx prey.

Keywords: critically endangered, main prey, recovery model, baseline survey

Status and conservation of the critically endangered balkan lynx - *The status of the Balkan lynx in Albania*

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The Balkan lynx *Lynx lynx balcanicus* is the most endangered autochthonous population of Eurasian lynx *Lynx lynx*. It is distributed in Albania and Macedonia and occurs also in western Kosovo. In the early 20th century, the Balkan lynx was present in all mountains and forests in Albania. However, continuous persecution and habitat destruction reduced its abundance and distribution in the second half of the 20th century. Before 2006, there had been a limited knowledge on distribution, abundance, biology and ecology of Balkan lynx in Albanian literature. Within the frame of the Balkan Lynx Recovery Programme we conducted a camera-trapping survey in 9 areas of Albania between 2009 and 2015. The first picture of the Balkan Lynx in Albania taken by camera – trapping survey was in Munella Mountain on March 26th, 2011. The results showed that lynx occur on Munella Mt. and its surroundings in central north Albania. From the identification of individuals by comparing 66 Balkan lynx pictures collected, a minimum of 4 mature individuals were identified in Munella. Finding of two dead lynx cubs in 2015, in Munella region, proved that this region is the only other area besides Mavrovo National Park in Macedonia where a small sub-population of Balkan lynx survives.

Keywords: Balkan lynx, *Lynx lynx balcanicus*, status, conservation, distribution, abundance, Albania

First insight into the spatial and feeding ecology of the critically endangered Balkan lynx (*Lynx lynx balcanicus* Bureš, 1941)

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In light of the large carnivore recovery in Europe, the Balkan lynx (*Lynx lynx balcanicus* Bureš, 1941) is facing an increased risk of extinction. Solid measures for its recovery need to be based on robust, up-to-date knowledge in order to tackle the main threats. Here, we present results of the first radio tracking study of the critically endangered Balkan lynx. Four male lynx were radio-tagged in the western part of the Republic of Macedonia using GPS/GSM collars. We calculated home-range size with traditional estimation methods (minimum convex polygon [MCP] and kernel density estimation [KDE]), and calculated habitat selection with Jacobs Index. The mean home-range size was 425 ± 151

km² for MCP and 574 ± 237 km² for KDE with reference bandwidth. The Jacobs Index revealed for second order habitat selection (i.e., selection of home ranges within the study area) strong variability between individuals, however, for third order habitat selection (i.e., selection within home ranges) Balkan lynx preferred forests and areas with sparse vegetation, and avoided agricultural areas and pastures.

Our study demonstrates that the knowledge of the individual home-range sizes, site fidelity and habitat preferences of the Balkan lynx can enhance the decision making process on management actions relevant to wildlife. Furthermore, knowledge about diet composition helps the management bodies account for ample amount of preferable prey that can support the existing and increasing population of the Balkan lynx. We believe that this study will contribute towards better management practices of the wildlife in Macedonia and open the door to new conservation-based research.

Keywords: Balkan lynx, home-range, GPS/GSM collars, conservation measures

Public perceptions of large carnivores in Albania and Macedonia

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We tested differences in attitudes towards bears, wolves and lynx among the rural public in Albania and Macedonia through information collected from a quantitative field survey between April 2007 and January 2009 (n = 759). Wolves were the species with the least positive attitudes among the rural public and had the lowest support for conservation compared to bears and lynx. In addition, conflict perception towards wolves was higher than for bears and lynx. Attitudes towards large carnivores seem to be influenced by respondents' factors related to country of residence, knowledge, education, gender, livestock ownership, damages experienced, and interest in hunting and hiking. We argue that, based on differences in public attitudes, conservation initiatives and management plans for large carnivores should keep wolves separate from bears and lynx as lower public support for wolves might jeopardise the conservation of the two other large carnivores. All three species need to be addressed separately from a conflict-management point of view.

Keywords: large carnivores, human–wildlife conflict, human dimensions, Albania, Macedonia

Distribution, numbers and density of the wildcat (*Felis silvestris* Schr.) in NP Vitosha, Bulgaria

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The European wildcat is an elusive animal which makes its study difficult. The species is Endangered according to the Red Book of Bulgaria and is protected by the Bulgarian Biodiversity Act. Its distribution, numbers and density are poorly studied in the country. The objective of this study was to collect data for the wildcat distribution and population size on the territory of Vitosha Nature Park, Bulgaria. 30 camera traps were moved between 6 predefined zones according to a 600x600 m grid in 2013 and 2014. The collected data were processed with Camera Base 1.6 and GIS instruments. The results of our study show that the wildcat is unevenly distributed in NP Vitosha due to several factors. NP Vitosha is frequently visited by people with or without domestic dogs. The network of settlements situated around the park contributes to the high number of feral dogs and cats - a direct threat to the population of the wildcat. These cause the absence of the species from the areas closest to the capital Sofia. The highest density of the wildcat is found in the southern part of the park which is less populated by humans. Our density estimations are similar to those reported for other European countries. The results of our study indicate that wild cats avoid areas with intense presence of humans and dogs. The presence of feral cats is a direct threat causing hybridisation. Hybrids were occasionally recorded during the study.

Keywords: cameratrapping, density estimation, threats

GPS telemetry of feral dogs (*Canis familiaris*, L.) in NP Vitosha, Bulgaria – behavior and conservation issues

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The feral dogs' behavior is poorly studied, although numerous studies report their significant impact on wildlife. The intensive camera trap studies and park rangers' observations in NP Vitosha, Bulgaria revealed considerable feral dog presence even in remote places away from humans. The aim of our study was to provide detailed information on the feral dogs' movements and behavior in the park

in attempt to understand their impact on wildlife. Four dogs were captured in the forested areas of NP Vitosha and fitted with GPS/GSM collars. The resulting data was analyzed with GIS instruments. One of the collars was destroyed by a human who claimed ownership of the captured dog. Another dog was killed by a rival dog pack. The remaining two dogs (female and male, both castrated) belong to one pack with core area located on the outskirts of Zheleznitsa village. The analysis of the gathered data revealed the dogs' maximum distance covered per trip (varies by season, between 16,9-25,8 km), home range (28-31 km²) and core area size (0,64 km²). The trips in the mountains outside of the core area usually start at dusk and the dogs return early in the morning. All of the frequently visited places are inhabited by abundant ungulate populations, which suggest that the dogs are attempting to hunt. The dogs use diverse food sources like garbage, food provided by tourists and food offerings left at the graveyard of the village. The telemetry data implies that the feral dogs have adaptive strategy, utilizing all accessible food sources – provided directly or indirectly by humans during the day and the available wildlife in the park during the night. Their prey items in NP Vitosha also include endangered species like the Balkan chamois (*Rupicapra rupicapra balcanica*).

Keywords: feral dogs, GPS/GSM telemetry, chamois, home range, activity

First data on the isolated Balkan population of Goosander *Mergus merganser* at Lakes Prespa and Ohrid

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The small, isolated population of Goosander *Mergus merganser* population at the transboundary Prespa Lakes and Lake Ohrid in the Balkans has not been studied extensively so far. The objective of this study was to collect data on the breeding status of this species. Censuses were conducted by one observer in a boat moving slowly along predetermined routes, around 100–150 m from the rocky shore. There were at least two coordinated censuses per year, from 2011 to 2015, one in mid/late April to early May to count courting pairs and prenuptial concentrations, and one in late May/early June to count females with young. It is estimated that there are 15–25 breeding pairs at Prespa Lakes, with a minimum population size of 77 individuals present in the breeding period. During the last twenty years, the Prespa Lakes have also been the most important wintering site for this population, holding from 4.8–100% of the total numbers of Goosanders wintering in the southwest Balkans (i.e. in Albania, the FYR of Macedonia and Greece). Up to 130 birds overwinter in the region, where numbers have increased during the last 30 years in both Prespa and Ohrid Lakes. A 2014 census confirmed the first evidence of 3–4 pairs of this species nesting at Lake Ohrid, the second most important wintering site in the region. Conservation of this isolated population is of utmost importance, at least in a European context.

Keywords: Goosander census, FYR of Macedonia, Albania, Greece.

Distribution, nest site selection and breeding success of White Stork in Republic of Macedonia

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During the national White Stork census of 2015 we selectively covered the territory of Macedonia (25.713km²) and recorded 817 pairs (HPa) during 2015 and another 17 pairs (HPa) in 2016 in a region not covered in 2015. The census was conducted in an attempt to establish a snapshot view into the White Stork population in Macedonia as well as a baseline for further conservation action. Most of the data was collected by volunteers with moderate training and their data while reliable was not always robust enough. The territory of Macedonia has 27 breeding colonies and only 5 of these colonies have more than 20 nests. The ecological density is 3.2 pairs / 100 km² but it varies significantly regionally. Since the census was done well into the breeding season (end of May to late July) we also have relevant data on pairs with fledged young (726 HPm) and breeding success of 2.47 fledged young per active nest which is a significant decrease in comparison to 3.16 in 1958. The survey also determined that there is a change in nest site selection from the last national stork census conducted in 1958; nesting on overhead transmission pylons while not present at all in 1958 is now the predominant nesting selection with 62,6%; nesting on buildings has decreased from 55,2% to 36,8%; nesting on trees has decreased even more significantly from 37,2% to 0,6% and while nesting on hay bales and stacks was present with 7,5% in 1958 it is now completely abandoned.

Keywords: breeding colonies, ecological density, white stork census

Faunistic data of Araneae from Skopje and Malesh valleys of R. Macedonia

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Data concerning Araneae of the Skopje and Malesh valleys in R. Macedonia are presented. The research underlying this study was carried out in the period of April-August 2014, at six localities in the Skopje and Malesh valleys, by using pitfall traps. A total of 100 species of the suborder Labidognatha belonging to 19 families and 55 genera were registered. Seven species, *Hyposisinga sanguinea* (Lathys humilis), *Trachyzelotes adriaticus*, *Zelotes harmeron*, *Zora silvestris*, *Episinus maculipes* and *Titanoeca*

quadriguttata are new for the Macedonian spider fauna, while three species, *Harpactea samuili*, *Zelotes babunaensis*, *Zodarion ohridense* are endemic for the Balkan fauna. The presence of seven new species, which were already confirmed for the surrounding countries, is expected due to the relative lack of faunistic data of Araneae in both areas. The relatively high number of registered species, as well as the presence of new ones, emphasises the need for further research.

Keywords: spiders, new species, R. Macedonia

Riparian habitats as hot spots for Orthoptera diversity in karstic environments

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Riparian zones represent highly productive and species rich habitats under strong influence of streams, which provide them with nutrients, ensuring elevated taxonomic richness when compared to adjacent terrestrial habitats. In a research conducted from May to November 2014, soil arthropods were sampled along Krčić River, a temporary river in south-eastern Croatia, both in the riparian zone and the surrounding karstic area. Sampling was performed along the entire flow using pit-fall traps, set in 72 stations in groups of three. From the samples thus collected, all grasshopper and cricket taxa (Orthoptera) were isolated and determined to the lowest taxonomic level possible (35 taxa and 1186 specimens in total), in order to perform community analyses and see how community structure differs between riparian and karstic habitats. Taxon richness and diversity were significantly higher in riparian than in karstic habitats, probably due to more diverse vegetation structure and favorable microclimatic conditions. Similar pattern was observed for activity density, which showed significantly higher values in riparian habitats. Also, karstic habitats cluster together with a high degree of similarity, containing a few dominant species such as *Calliptamus* sp., *Gryllomorpha dalmatina* and *Mogoplistes brunneus*. It can be concluded that riparian zones are indeed very important in harbouring Orthoptera diversity in karstic regions and as such should be especially valued in terms of nature protection and species conservation.

Keywords: Krčić River, community analyses, species richness, activity density, conservation.

Synthesys Collections Self-Assessment Tool – European Standards for Natural History Collection Management

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The SYNTHESYS is a European Union- founded program running for more than 10 years. It is split up for four-year long projects. In the current project 20 European natural history museums, universities, botanic gardens, and research organisations participate.

The SYNTHESYS has already been well-known about its Access part, which offers opportunity the researchers to visit the larger European natural history collections. The main aim of the project, however, is to produce an accessible, integrated European infrastructure for the research users and natural history collection.

The SYNTHESYS has created an online Collections Self-Assessment Tool (CSAT) for natural history collection holding institutes to help them identify the critical areas of their collection management. By gaining valuable information from the questions and advice that the forms provide, the institutions may have a better knowledge of European standards. Our museum has a mission to involve the natural history collections of the Balkan and Carpathian Basin areas by encouraging them to fill the CSAT questionnaires on the website that we specifically created for this purpose.

Keywords: Synthesis, Collections Self-Assessment Tool

Contribution to the exploration of the bryophyte flora of the Republic of Macedonia with a special attention to the species of conservation interest

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The Republic of Macedonia is one of the bryologically poorly explored countries in Southeast Europe. During the last five years short field trips added a lot of new species to the Macedonian bryophyte flora; 82 new national records (26 liverworts and 56 mosses). Hence the bryophyte flora of the country at present consists of 573 taxa (106 hepatics and 467 mosses). The number of the known bryophyte taxa is still about 200–250 less than that of the neighbouring countries (Bulgaria: 807, Greece: 723, Serbia: 723). Only Albania has a lower number of bryophyte taxa (466), which is also due to the under-explored bryoflora of this country. On the other hand, many bryophytes have only one or two records from the Republic of Macedonia, hence there is not enough knowledge to evaluate the threat status of species in the country or to establish a list of important bryophytes from a conservation point of view. However the number of species of European conservation interest is high. 22 species are included in the Red data book of European bryophytes. Further 49 species are on the candidate list of the new Red data book of

European bryophytes and a high number of species can be regarded as rare on the Balkans.

We will give an overview on the recent knowledge of the conservationally important bryophyte species in the country with special attention to their ecology and the threatened habitats.

Keywords: Bryophyta, Macedonia, species of conservation interest

Current stage and future perspectives of diatom diversity research in Macedonia

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The diatom research in Macedonia started at the beginning of the 20th century, with a focus on diversity in ancient lakes Ohrid and Prespa. By the mid 20th century, Lake Ohrid as renewed ‘biodiversity hot-spot’, took the research priority. Intensive taxonomic research resulted with a record of ~400 diatom taxa, 75 of which were new for the science. Later on, research interests broadened towards Lake Dojran, River Vardar and its tributaries and mountain wetlands. As result of these studies, the number of recorded diatom taxa increased up to more than 630. At the beginning of the 21st century the diatom research was intensified and expanded on various freshwater habitats. Two research approaches were established: 1) investigations of the regional flora, and 2) detailed revision of genera or species complexes. The first approach was undertaken in frame of various valorization projects, conducted by the Macedonian Ecological Society (Osogovo, Jablanica, Bregalnica watershed, etc.), Biology Students Research Society (Shara, Kozhuf, Pelister, Korab, Karadzica etc.), or scientific projects (lakes Ohrid and Prespa). The second approach includes observations of historical samples deposited at the Macedonian National Diatom Collection (MKNDC) over the last century. As result, more than 120 new species were described within several genera, including *Achnanthes* sensu stricto, *Amphora*, *Cyclotella*, *Diploneis*, *Eunotia*, *Gomphonema*, *Hippodonta* and *Luticola*. Moreover, the continuous research on lakes Ohrid and Prespa, revealed existence of additional 100 new taxa for the science. This comprehensive research yielded with a record of more than 1400 diatom taxa for Macedonia. The ongoing and future research will follow the same approach. In this regard, the studies will focus on high-mountain and specific habitats as thermo-mineral springs, halomorphic soils, subaerial habitats, calcareous springs etc. In addition, several species-rich genera as *Caloneis*, *Neidium*, *Nitzschia*, *Pinnularia* and *Sellaphora* need to be re-examined. It is expected that the diatom flora of Macedonia will count around 1800 species which covers around 65% of the European flora.

Keywords: diatom diversity, Macedonia, regional flora, evaluation

First records of *Polycentropus irroratus* Curtis 1835 and *Wormaldia juliani* Kumanski, 1979 (Insecta, Trichoptera) for the fauna of Republic of Macedonia

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Trichoptera is a very rich order in the Balkan Peninsula, but knowledge of caddisflies in the Republic of Macedonia is still not complete. Adult caddisflies were collected during June 2016 on the Orevoevka Reka River, after the local fishpond, using ultraviolet (UV) light traps and an entomological hand net. Herein, *Wormaldia juliani* Kumanski, 1979 and *Polycentropus irroratus* Curtis 1835 are reported for the first time for the Macedonian caddisfly fauna. The Balkan endemic *W. juliani* has restricted distribution in the neighboring countries (Greece and Bulgaria), while *P. irroratus* is widely spread caddisfly in European countries. We also provide notes on ecological aspects and taxonomical remarks on some species. The recent discovery of a new species presents important contribution to the knowledge on the rich freshwater biodiversity in R. Macedonia.

Keywords: Trichoptera, *Polycentropus irroratus*, *Wormaldia juliani*, Orevoevka Reka River, R. Macedonia

An overview of the macro-crustacea (Invertebrates-Arthropoda) in Special Nature Reserve Zasavica

Mihajlo Stanković

Pokret gorana-SRP Zasavica

For the past eighteen years of research Fauna Reserve Zasavica total recorded 20 species Macrocrustacea. Subclass Branchiura the reserve is represented with Fam: Argulidae and ichthyophagic parasitic species *Argulus foliaceus* from the subclass Malacostraca ordo Isopoda present cosmopolitan species *Asellus aquaticus* In the reserve was discovered five of Amphipods, of which three are surface modes (*Dikerogammarus fluviatilis*; *Gammarus balcanicus* and *Gammarus fossarum*), and one semi-underground type (*Synurella ambulans* underground type *Niphargus valachicus*). Of these Amphipoda *Niphargus valachicus* is a strictly protected species in Serbia. Total discovered three types of branhipods and they are: *Chirocephalus brevivalpis*, *Lepidurus apus* and *Cyzicus tetracerus*. *Chirocephalus brevivalpis* is a rare species, known only from a few localities in Romania and in two

areas in Serbia, a report on *Zasavica* is the westernmost of this kind and the only one south of the Sava and Danube rivers. The species *Lepidurus apus* with very limited distribution in Serbia, found only in two locations in central Banat. Finding kind in Batar tributary is unique because the individuals were found in running water. The Reserve has so far found nine species of ostracods and they are: *Candona aff. candida*, *Cypria ophthalmica*, *Cycloprsis laevis*, *Cycloprsis ovum*, *Eucypris virens*, *Bradleystrandesia reticulata*, *Heterocypris incongruens*, *Cypris puber*, and *Ilyocypris sp.* Although widespread and common species in the Euro-Asian continent, with exceptional tolerance to different habitat conditions record of *Candona aff. candida* in *Zasavica* is the first in Serbia. From the ordo Decapoda and Fam. Astacidae in *Zasavica*, *Astacus leptodactylus* was found, that was introduced to Central Europe in the nineteenth century from the Caspian Sea region and can grow up to 30 cm in length. Type *Astacus leptodactylus* favors relatively calm waters (lakes, ponds, marshes and canals) and is located on the IUCN Red List.

Keywords: makrocrustacea, *Zasavica*,

Distribution of decapod crustaceans (Decapoda: Potamonidae, Astacidae, Atyidae) in the Republic of Macedonia: conservation approach

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Five decapod crustaceans occur in freshwater habitats in R. Macedonia: noble crayfish *Astacus astacus*, stone crayfish *Austropotamobius torrentium*, freshwater crabs *Potamon fluviatile* and *P. ibericum*, and freshwater shrimp *Atyaephyra stankoi*. Despite the importance of decapod fauna as keystone species in aquatic habitats, insufficient information on the distribution of this indigenous species from the territory of R. Macedonia is available at present. Additionally, crayfishes, crabs and shrimp, like the freshwater habitats in which they are encountered, receive relatively little publicity and conservation attention in the country, in spite of their key role in aquatic food webs and ecosystem functioning. This study (i) critically review the historical data on the occurrence and distribution of individual representatives of the decapod species; (ii) summarizes the data found in the collection of the decapoda in the Macedonian Museum of Natural History (MMNH) Skopje, R. Macedonia and (iii) present unpublished records of the crayfish in the country by the first author. The paper contributed to update our knowledge on the geographical distribution, ecology and taxonomy of the freshwater decapods in R. Macedonia and will serve as a valuable source of information for nature conservation and the protection of decapods and their habitats.

Keywords: decapod crustaceans, distribution, conservation, R. Macedonia

New occurrence and ecological features of *Tinodes janssensi* Jacquemart, 1957 (Insecta, Trichoptera) from the Republic of Kosovo

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A rare caddisfly *Tinodes janssensi* Jacquemart 1957 is endemic species of the Balkan Peninsula and has been rarely reported outside Greece. In this contribution we report new occurrence of this species in the Republic of Kosovo, more precisely from Kishnapole village, in the Municipality of Gjilan. This is the third known locality of this species in Kosovo. Outside Greece, from where it was described, it is only known from a single locality in Albania and only few localities in Bulgaria. In all three localities in Kosovo, *Tinodes janssensi* is characterized as a subrecent species with only few specimens. Even though from Greece and Bulgaria it has been reported to fly from April to November, in Kosovo it has only been found during May, June and July. This species in Kosovo is found in upstream areas of small streamlets, inside or close to the forested areas and outside anthropogenic impact.

Other genera associated with *Tinodes janssensi* in samples of the particular months are: *Rhyacophila* Pictet, 1834, *Hydropsyche* Pictet, 1834, *Oecismus* McLachlan, 1876, *Philopotamus* Stephens, 1839, *Wormaldia* McLachlan, 1865 and *Potamophylax* Wallengren, 1891.

Keywords: Caddisflies, Psychomyiidae, endemic species, rare species, Balkan Peninsula.

Diversity and distribution of Dragonflies (Odonata) in the river catchment of Bregalnica and their importance in defining the protected areas in East Planning Region of Macedonia

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This paper presents the results of a survey of the diversity and distribution of dragonflies (Odonata), with emphasis on important species and their specialization to the habitats in the river basin of Bregalnica. Different types of habitats (lotic and lentic water ecosystems) are covered from the source area to the water mouth of Bregalnica to river Vardar, including major tributaries of Bregalnica and certain specific habitats in the wider area. 41 species of dragonflies are registered, five of them have conservation importance (*Coenagrion ornatum*, *Caliaeschna microstigma*, *Ophiogomphus cecilia*, *Gomphus flavipes* and *Cordulegaster heros*). As a major result, several areas/localities are prioritised for protection and as a part of the proclaimed and proposed protected areas particular importance for the

conservation of dragonflies have: *Dolna Bregalnica, Mokra Livada-Kukuljeto, Zrnovska Reka, Ratkova Skala - Zletovska Reka and Dolna Zletovica.*

Keywords: dragonflies (Odonata), conservation, Bregalnica

Contribution to the fauna of butterfly family (Lepidoptera: Sphingidae) in the Republic of Macedonia

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Research of literature data, data from Macedonian Museum of Natural History collection, collection of Museum Nikola Nazlibinski – Struga and collection of Tobacco Institute in Prilep, showed that this family of Butterflies (Lepidoptera: Sphingidae) is well investigated in qualitative terms. If we take into account the numeral representation of Macedonian fauna species compared to Europe, out of 40 known species in Europe, Macedonia confirmed 25 species hawk moths. Three Lepidoptera (Sphingidae) species are new for the fauna of Macedonia: *Hyles gallii* (Rottemburg 1775), *Hyles nicaea* (de Pruner 1798) and *Sphinx pinastri* Linnaeus, 1758. According to Von Nieuwerkerken et al. (2011), 2 species: *Theretra alecto* (Linnaeus, 1758) and *Sphingoneopsis gorgoniades* (Hübner, 1819) are indicated for the presence in Macedonia, and for them there are no reference data in literature available to us. But any new future species data or localities will upgrade the image of their species representation and distribution in Macedonia.

Keywords: faunistics, new species, Lepidoptera: Sphingidae, Republic of Macedonia

Preliminary investigation of the distribution of *Batrachochytrium dendrobatidis* in Serbia

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In the past decade, amphibians across the world have experienced dramatic reductions in population densities. The causes of these declines are numerous and often have devastating consequences. Diseases

are a substantial cause of mortality of amphibians worldwide. One of today's most prominent and least studied disease is caused by the pathogenic fungus *Batrachochytrium dendrobatidis* (*Bd*).

In 2015, we collected 88 amphibian skin swabs from 12 localities in the Republic of Serbia. The samples came from 7 species: *Bombina orientalis*, *Bufo bufo*, *Ichthyosaura alpestris*, *Rana dalmatina*, *Pelophylax ridibundus*, *Pelophylax lessonae*, and *Pelophylax esculentus* and the sites ranged in altitude from 67 m (a.l.s.) to 930 m (a.l.s.).

Overall, 7 samples (8%) were positive for *Bd* and belonged to the species *Pelophylax lessonae* (n = 4), *Pelophylax kl. esculentus* (n = 2), and *Pelophylax ridibundus* (n = 1). *Bd* was found at 4 localities belonging to federally protected nature reserves and unprotected sites. Within these localities, between 11% (locality Jarkovci) and 50% (locality Apatin) of amphibians were positive for *Bd*.

This preliminary research, conducted for the first time in the territory of the Republic of Serbia, indicates potential danger of this pathogen. Our future goal is to organize a detailed survey from a larger number of sites in order to determine the actual threat and degree of vulnerability.

Keywords: *Batrachochytrium dendrobatidis*, amphibians, diseases, presence, Serbia.

Diversity of intestinal helminths of the red fox (*Vulpes vulpes* L.) in Vojvodina (Serbia)

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Red foxes (*Vulpes vulpes*) are efficient predators, able to adapt to different habitats and food sources. Additionally, they are also important carriers and vectors of zoonoses. After successful oral vaccination against rabies, fox numbers have risen in many European countries including Serbia, with their ranges spreading into urban habitats. Contact with humans and domestic animals is becoming more frequent, increasing the probability of pathogen transmission.

The aim of this study was to determine the diversity of intestinal helminths, particularly zoonotic and anthrozoönotic parasites, in foxes in Vojvodina. Analysis was conducted on 32 foxes captured between October 2012 and May 2015. All of the animals were delivered to the Pasteur Institute in Novi Sad. The intestinal scraping technique and the Lörintz method of coprological analysis were used to detect parasites.

Of the examined foxes, 96% were infected with a total of 1156 individual helminths belonging to 9 species: *Alaria alata* (Trematoda); *Mesocestoides litteratus*, *Taenia pisiformis*, *Taenia hydatigena* and *Echinococcus multilocularis* (Cestoda); *Toxascaris leonina*, *Toxocara canis* and *Uncinaria stenocephala* (Nematoda). Tapeworms were the most prevalent helminths (81%), followed by nematodes (40%) and digeneans (9%). The most numerous helminth species was *E. multilocularis* (550 individuals), and the most prevalent was *M. litteratus*. *Toxocara canis*, *Uncinaria stenocephala* and *Echinococcus multilocularis* are potential agents of zoonoses.

Toxocara canis causes toxocarosis, and humans are potential paratenic hosts of this nematode. *Uncinaria stenocephala* causes ancylostomiasis in foxes and other carnivores, including domestic dogs. *Echinococcus multilocularis* causes echinococcosis, one of the most dangerous known

anthropozoonoses. This was the first finding of this tapeworm species in red foxes in Vojvodina, and the majority of infected animals are from the Vrdnik-Ruma-Irig area, which can be considered an infection hotspot. The presence of these species emphasizes the necessity of monitoring fox helminth fauna, as well as the application of preventive and protective measures.

Keywords: digeneans, tapeworms, nematodes, zoonoses, anthropozoonoses

Loss of key type of habitats and flora and fauna species in Vardar River Basen, Macedonia (1971-2015)

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The paper analyzes the spatial development plans and other documents relating to several regions in the Vardar basin, and the changes that have occurred in the past 45 years (1971-2015).

The data obtained from the analysis of selected documents and those obtained by field surveys in regions of the Vardar basin with sub-basins of rivers, Lepenec, Pchinja, Topolka, Babuna, Bregalnica, Crna Reka, Boshava, Anska Reka are presented. Changes of the ecosystems in the watershed are shown cartographically.

Keywords: habitat types, vegetation communities, flora, fauna, ecological corridor, Vardar river basin, Macedonia.

The Balkan impact upon Slovenian flora

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There are several delimitations of the Balkan peninsula (BP) from simplified line Triest-Odessa to rivers Danube-Sava-Kolpa or combination of northern borders of political entities, but the sharpest obstacle for the historical plant migrations in NW part of the BP is the E Alpine arc, with the range above 2000 masl somehow in NW Slovenia (and so with permanent glaciation during Pleistocene). In W Slovenia there is as small but clear-cut altitudinal gap between Dinaric and Alpine mountains somewhere around Postojna with only about 600 masl and in the area between SE Julian Alps and N Velebit in a distance of over 150 km there are only very few and scattered mountain peaks above 1500 masl. So migration of Balkan mountain flora to the N beyond Velebit should have been difficult, with several local extinctions and hence diversity of Balkan taxa in Slovenia is depauperate. Turrill reported in 1929 that more than 1/3 taxa (sub-) endemic to the BP and of those only a fraction is to be expected in the NW corner in contact to the Alpine flora. Studying Slovenian flora about 40 taxa turned out to

be the “real” Balkan with distinctly major part of the distribution range limited to the BP, about 20 taxa with Illyric distribution pattern and further 20 Illyricoid, following the combined pattern of Alpine-Illyric (25), Apennine-Illyric (10), Illyric-SE Alpine-Apennine (30) etc, in total about 240 taxa, which is about 10% of Slovenian native flora. Their occurrence is distinctly concentrated in SW Slovenia and in the southern outskirts of the Alps plus Zasavje (area E of Ljubljana along the Sava River). Locally they represent up to 20 % (30 %) of flora with higher proportion in geographically diverse areas. So definitely Slovenian flora has an important Balkan imprint and at least SE Slovenia is a part of the Balkan biogeographic region.

Keywords: flora of Slovenia, Balkan flora

Threatened bryophytes on Fruška gora Mountain (Serbia)

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Fruškagora is mountain located in the southern part of Pannonian plane. Considering that this area is mainly covered by forest vegetation, and characterized by dense hydrological network, bryophytes are often conspicuous elements of different vegetation types on this mountain. The knowledge on threatened and rare bryophytes is very important in order to preserve these species. Bryophyte flora of Fruškagora counts around 150 taxa (20 liverwort and 130 moss species). That number represent about 17% of Serbian liverwort, and 23% of moss flora. Total number of 13 (around 8% of total bryophyte flora on Fruškagora) species are threatened. One liverwort and 10 moss species are listed on Bryophyte red list of Serbia and Montenegro. Of that number, two species are “critically endangered”, two “endangered”, two “vulnerable”, three species are in “low risk” category, and one in “data deficient”. One moss species (*Pseudocampyllumradicale* is listed in Red Data Book of European Bryophytes (ECCB, 1995) in “rare” category. Four species are candidates for the new Red Data Book of European Bryophytes.

Complete bryophyte flora of Fruškagora is still unknown. Considering the fact that this is the region with high biodiversity, dense hydrological network, and very complex geology, there are a lot of different microhabitats which are conducive to some rare bryophytes.

Keywords: Bryoflora, Liverworts, Moss, Red list

Pondweeds (*Potamogeton*) in Croatia – recent status and ecology

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Monitoring of macrophytes in Croatian rivers and lakes in the period from 2009 to 2016 has resulted in comprehensive data set considering distribution and ecology of water plants. Among them, pondweeds (*Potamogeton*) are one of most frequent groups in macrophytic vegetation. Up to now, their recent distribution, affiliation to plant communities and ecological conditions of water where they occur, were very scarce or even completely lacking. Thus, a new insight into pondweeds of Croatia has been based on sampling on over 300 localities and on revision of existing data. For the flora of Croatia 20 taxa of *Potamogeton* are quoted based on existing data and recent studies, including five hybrids. However, some records are very old and uncertain, e.g. for *P. alpinus*, *P. compressus* and *P. polygonifolius* there are only single herbarium records from the mid-19th century. Some species, as *P. filiformis* are rare with single findings, and other have less than dozen known localities. Based on our field research, only six species have wider distribution (*P. nodosus*, *P. natans*, *P. pectinatus*, *P. lucens*, *P. perfoliatus* and *P. crispus*), enabling more reliable ecological analyses. They showed that e.g. higher abundance of *P. pectinatus* in streams is correlated with increased levels of nitrites, that occurrence of *P. crispus* indicates higher levels of nitrates; while *P. perfoliatus* and *P. lucens* are usually present in ecologically stable environment. Furthermore, two hybrid taxa new for the flora of Croatia were recorded – *P. × salicifolius* (*P. lucens* × *P. perfoliatus*) and *P. × angustifolius* (*P. gramineus* × *P. lucens*) – both on localities where parental taxa were present. Thus, to the extent of our knowledge, Croatian flora has rich pondweed flora, but with low number of commonly distributed species and some still doubtful records.

Keywords: macrophytes, water quality, aquatic vegetation

New and rare plant taxa for the flora of the Republic of Macedonia

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Floristic researches continually carried out on the territory of the Republic of Macedonia keep supplementing current data by adding new information concerning genera and species belonging to previously published families in the edition “Flora of the Republic of Macedonia”, as well as families that are yet to be published in the future volumes of this edition. This paper mentions the data regarding distribution of following new or rare plant taxa on the territory of the Republic of Macedonia, which had been discovered in the past period - *Andrachne teliphioides* (Štip-Isar) (new for the Flora of the Republic

of Macedonia), *Astragalus hypoglottis* subsp. *gremlii* (Jablanica-Vevčani and Podgorechko Ezero), *Bellardia trixago* (Prilep-Debreška Krasta; Mariovo-Sliva), *Campanula pichleri* [*Asyneumapichleri*] (Jablanica-Vevčansko Ezero), *Chorisporea tenella* (Skopje-Madžari) (new for the Flora of the Republic of Macedonia), *Dactylorhiza viridis* (Jablanica-Vevčansko Ezero), *Fumana arabica* (Valandovo), *Geranium versicolor* (Struga - Crn Drm gorge - v. Modrič), *Glinus lotoides* (Prilep-Mariovo, Crna Reka gorge, Rasim Bej Most), *Ilex* (Jablanica-Vevčani), *Kitaibelia vitifolia* (Jablanica-Vevčani), *Lagoecia cuminoides* (Krivolak), *Marrubium pestalozze* (Kriva Palanka-Psača) (new for the Flora of the Republic of Macedonia), *Nepeta parviflora* (Ovče Pole-Vrsakovo, Sudič) (new for the Flora of the Republic of Macedonia).

Model for monitoring of biodiversity in protected areas in Bosnia and Herzegovina

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The heterogeneity of Bosnia-Herzegovina, which includes the geomorphological history and ecoclimate diversity, have caused the especially rich living world in the area. The flora, fauna and fungia of our country are deemed to be one of the most diverse in the whole of Europe. High degree of endemism and relictiness gives them high position on the level of global biodiversity. Despite these facts, Bosnia-Herzegovina is a country with the smallest coverage of protected areas in Europe. The number and size of protected areas is slowly increasing in recent years.

The process of establishing a protected area, as an initial step, involves an evaluation of biodiversity. Detailed field and laboratory studies are conducted in order to assess the conservation status of species and ecosystems. In order to effectively protect biodiversity, in some of the designated protected areas the process of monitoring the status of endangered species and ecosystems was established. Results of biodiversity evaluation in a given area were used as the baseline.

Over past years, along with development of institutional capacities, one model for monitoring the status of biodiversity was prepared. By using modern software (Map Info Professional Turboveg, Juice, BiodiversityPro) indicators of biodiversity have been defined. The model consists of a two-stage monitoring. The first stage involves monitoring of phytocenose's structure and monitoring of pressures. The second level involves monitoring of ecosystem's functions. Adapting the suggested model to local specificities, makes it possible to track the intensity of anthropogenic pressures, eventual spreading of invasive species, the impact of natural disasters and other events. For the purposes of this paper, as a case study two protected areas in Canton Sarajevo (Monuments of nature Vrelo Bosne and Skakavac) have been selected. The proposed model can serve as a tool for monitoring of biodiversity in all protected areas of Bosnia and Herzegovina.

Keywords: model, monitoring, biodiversity, conservation ecology, anthropogenic pressures.

Protected areas network in Macedonia – are we doing enough to reach CBD Aichi Target 11?

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At the moment, 86 protected areas in the Republic of Macedonia are covering about 9% of the country territory however with uneven distribution, mainly covering the western part of the country (forests and lake ecosystems).

Republic of Macedonia committed to contribute towards Aichi Target 11 of the Convention on Biological Diversity that calls for protection of at least 17 % of terrestrial and inland water and 10 percent of coastal and marine areas to be achieved by 2020.

Despite the priority directions given in the National Biodiversity Strategy and Action Plan there is no significant progress in the area coverage of protected areas and the 'quantitative aspect' is far from reaching by 2020 although the National Spatial Plan goal for protection of about 12% of the county territory was adopted in 2004.

Information regarding 'areas important for biodiversity' at least with respect to 24 Important Bird Areas and 42 Key Biodiversity Areas exist however almost without any protection (about 80% of these areas are outside the national protected areas network). 'Management effectiveness' (for at least another 30% of protected areas) is the most difficult to be reached given the fact that management authorities have been established only for national parks and several other protected areas, some of them with very few activities in place. Integration of protected areas into wider landscapes and other effective area based conservation measures is not available.

Without acceptance of updated national biodiversity plan and commitment of financial resources for implementation of 2020 Aichi Biodiversity Targets, it appears highly unlikely that Macedonia will achieve its international commitment.

Keywords: Convention on Biological Diversity, Aichi Biodiversity Target 11, Management effectiveness indicator, protected areas

Larval description of *Drusus sharrensis* Ibrahimi, Vitecek & Previšić (Insecta, Trichoptera) endemic species of Sharr Mountains

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The Balkan Peninsula is one of the most interesting centres of diversity for different animal groups in Europe including Trichoptera. In this region about 45 species from the Eurasian genus *Drusus* (family Limnephilidae) have been recorded. In this study we present morphological and ecological features of the last instar larvae of *Drusus sharrensis* with data about distribution of this species. We also included the most important diagnostic features enabling separation of larvae of *D. sharrensis* from larvae of the other European Drusinae and Trichoptera species. Additionally, information on the sympatric caddisfly species of the springs where larvae and adults of *D. sharrensis* were found is presented. *Drusus sharrensis* is endemic species of Sharr Mountains.

Keywords: Caddisfly, Drusinae, southeast Europe, larval description, fauna, Republic of Macedonia

Distribution of the Genus *Cernosvitovia* (Oligochaeta, Lumbricidae) on the Balkan Peninsula

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Cernosvitovia is a genus with archaic characteristics. The genus is presented with 11 species on the Balkan Peninsula: *Ce. biserialis*; *Ce. bulgarica*; *Ce. dobrogeana*; *Ce. munteniana*; *Ce. rebelii*; *Ce. schweigeri*; *Ce. crnicae*; *Ce. dudichi*; *Ce. getica*; *Ce. Krainensis* and *Ce. opisthocystis*, most of them endemic to the region. The species *Ce. crnicae* and *Ce. dudichi* are endemic to Serbia, *Ce. bulgarica* and *Ce. biserialis* – endemic to Bulgaria, *Ce. munteniana* – endemic to Romania and *C. shweigeri*, is endemic to north-western Turkey.

The objective of our study is to analyse the whole list of literature records in order and to present a general overview of the distribution of *Cernosvitovia* genus on the Balkan Peninsula.

Keywords: Cernosvitovia, Earthworms, Annelida, Lumbricidae, Balkan Peninsula.

The earthworm populations in National Park Persina, Bulgaria

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In 2015 and 2016 were investigated the soil-dwelling annelids in Persina, one of the newest National parks in Bulgaria. Earthworms were sampled by digging and hand-sorting the soil. The data of the Lumbricidae species composition on the following areas of the park were studied. Four species *Allolobophora leoni*, *Octolasion lacteum*, *Dendrobaena veneta* (& *Eisenia fetida*) belonging to four different genera were established. Some biocenosis indicators as dominance, and frequency were considered as well. Of 4 species only *O.lacteum* present at all sites, occurred in large numbers.

Keywords: earthworms, species diversity, dominance, frequency, National park, Persina, Bulgaria

First record of black bullhead *Ameiurus melas* (Pisces, Ictaluridae) in R. Macedonia

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The North American catfish – the black bullhead, *Ameiurus melas* is recorded for the first time in R. Macedonia, in Pcinja River. Nine individuals of this ictalurid fish species were caught from the anglers in Pcinja River on September 2015, another 8 samples on May 2016 and brought to the Institute of Biology, Faculty of Natural Science and Mathematics in Skopje. After a detailed observation of the external morphological features, osteological architecture, especially of the pectoral girdle and pectoral spine, it was concluded that the individuals had all the distinguishing features of the species *A.melas*. The presence of different size class permits the supposition that the black bullhead has established a self-sustaining population in this river.

The origin and introduction pathway of the black bullhead to the Pcinja River remain unclear, but there are three possible scenarios: (1) probably due to natural spread of individuals from Serbian populations; (2) this species exist in R. Macedonia as long as *Ameiurus nebulosus*, but because both species are very similar, the misidentification of black bullhead for such a long time is not surprising; (3) presuming the second scenario is true, then it is possible that the black bullhead was accidentally introduced with the brown bullhead in reservoirs containing the offspring of carp intended for breeding in the cage farms. The most likely period is before 2008, when regulations regarding imported fish from Serbia were poorly implemented or non-existent allowing *A. melas* to be illegally transferred to Pcinja River by anglers.

As many studies in Europe showed that distribution of *A. melas* in the water bodies is related to the fading of *A. nebulosus*, it is necessary to undertake urgent surveys to ascertain the current distribution and the ecological impacts of this invasive species in Macedonian waters.

Keywords: black bullhead, *Ameiurus melas*, Ictaluridae, catfish, Pcinja River, alien species.

First Record of *Eudiplozoon nipponicum* Goto, 1891 (Monogenea) in the Sava River, Bosnia and Herzegovina

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During the research period from January to July 2016, 200 fish individuals from the Sava River were collected and parasitological examined. Aim was to reveal ectoparasitic species. Fish sample was collected using fishing nets in cooperation with commercial fishermen. Parasitological analyze was conducted using microscopic technique and morphometrical parameters. As a result of investigation, 8 individuals of *Eudiplozoon nipponicum*, a monogenean trematode species was collected from the gills of *Barbus barbus*, *Abramis brama* and *Blicca bjoerkna*. This is the first record of this monogenean in the Sava River in Bosnia and Herzegovina. This gill monogenean is recorded in Hungary carp fish farms and in France. Also, it was detected in fish species collected from Czech Republic and France using combined morphological and molecular approach. *Eudiplozoon nipponicum* origin from Japan and after introduction with amur wild carp and carp a new habitat became Russia and Central Europa.

Keywords: *Eudiplozoon nipponicum*, Sava River, Bosnia and Herzegovina

Contribution to the study of spiders (Araneae) in Shar Mountain, NW-Macedonia

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Despite the fact that Shar Mt. is one of the highest (Titov Peak, 2748 m a.s.l.) and according to the surface (840.2 km²), one of the largest mountains on the Balkan Peninsula, from arachnological point of view it still remains unexplored. One of the main reasons for such gaps in the knowledge is the sporadic research and complicated history of the area. So far, information about the spider fauna of Shar Mt. can be found in four papers only, of which the work of Blagoev (1999) and Komnenov (2002) are especially devoted to the Shar Mt. In this study, information on new records and total number of species occurring in the area is given. The new records are discussed from faunistical and zoogeographical point of view. Comparison of the composition and richness of the spider fauna of Shar Mt. with other neighboring mountains is discussed, as well.

Keywords: Shar. Mt., spider fauna, new records

Recent findings of nectriaceous fungi (*Hypocreales*) in Bulgaria

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Nectriaceous fungi (*Nectriaceae*, *Hypocreales*) comprise 646 species from nearly 57 genera - parasites and saprobionts. Most of the members of this group often are treated as phytopathogenic organisms. Some of them are thought to be rare species in Europe and America, and undiscovered new ones are still expected to occur.

In this study, our findings from Northeastern Bulgaria, Forebalkan, Stara Planina and Rila Mts will expand the present data on the distribution of the nectrioid fungi in Bulgaria.

All specimens, gathered during the field studies carried out mostly in the period of 2015-2016, are conserved at the Mycological Collection of the Institute of Biodiversity and Ecosystem Research, Bulgarian Academy of Sciences, Sofia (SOMF). The microscopic features are studied with the aid of LM in water, 5% KOH and lactophenol in Cotton Blue. Color photographs are taken by means of Olympus and Canon PS digital cameras under Olympus BX-41, Boeco BM-117, Boeco-180/T/SP LM and BOE3500 dissecting stereomicroscope.

Cosmospora magnusiana is recorded for the second time from Bulgaria on fungal stromata on *Acer* twigs in Forebalkan. *Pseudocosmospora eutypellae* is confirmed with fresh collection from one more new locality in Stara Planina Mts. *Neonectria coccinea* is recorded for the first time from the Forebalkan, while *Nectria cinnabarina* is confirmed for the first time with sexual morph in Rila Mts and with non sexual morphs in Northeastern Bulgaria and Eastern Forebalkan. Brief descriptions, color illustrations and notes on their known distribution in the country are included.

Keywords: Cosmospora, fungal diversity, Nectria, Neonectria, Pseudocosmospora

New and interesting records of *Ascomycota* in Bulgaria

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In this study, our recent findings from Forebalkan, Vitosha region and Rila Mts will expand the present data about the fungal diversity and the distribution of the pyrenomycetous fungi from orders *Xylariales* and *Diaporthales* in Bulgaria.

The cited specimens, collected between 2015 and 2016 years, are conserved at the Mycological Collection of the Institute of Biodiversity and Ecosystem Research, Bulgarian Academy of Sciences, Sofia (SOMF). The microscopic features were examined in water and in solution of lactophenol in Cotton Blue under LM. Measurements of the microstructures were taken with the help of specialized software Carnoy 2.0. The color photos were taken with the aid of digital cameras under Olympus BX-41, Boeco BM-180/T/SP LM and BOE3500 dissecting stereomicroscope.

Anthostomella cornicola on dry bark of *Fagusa sylvatica* from Rila Mts (Rilomanastirska Gora Reserve), *Diatrype bullata* on dead twigs of old *Salix* tree (Ibar Reserve) from the Xylariales, and

Ophiognomonia melanostyla (Diaporthales) on dead leaves of *Tilia platyphyllos* (Vitoshka region, Nature Park Vitoshka), are reported for the first time in Bulgaria. *Gnomoniella tubaeformis* (Diaporthales) represents a new record from Vitoshka Mt. and the second find in our country. Its host-plant, *Alnus viridis* appears as the first report from Europe of *G. tubaeformis*. *Gnomoniopsis comari* s. lat. has never been recorded before on *Filipendula vulgaris* from Bulgaria. Brief descriptions, based on the examined specimens and data on the distribution of these pyrenomycetes throughout the country are given.

Keywords: Anthostomella, Diatrype, new host-plants, Ophiognomonia

Taxonomy, Distribution and Ecology of the Genus *Amylostereum* Boidin (Basidiomycota, Fungi) in the Republic of Macedonia

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This is the first paper of systematic research of the genus *Amylostereum* Boidin in the Republic of Macedonia. The survey for *Amylostereum* in this research area up to now revealed all three species known in Europe: *Amylostereum areolatum* (Chaillat ex Fr.) Boidin, *A. chaillatii* (Pers.) Boidin and *A. laevigatum* (Fr.) Boidin. According to the recent studies of their distribution in Macedonia, the species *Amylostereum areolatum* is a very rare one and is known only as a saprotroph on spruce trees, *Amylostereum chaillatii* is more common species registered mainly on fallen branches on fir, while *Amylostereum laevigatum* is known from several localities on different juniper species. The species of this genus are symbiotic fungi of woodwasps (Hymenoptera: Siricidae), which cause ecological and economic important damages. Within this paper is provided a brief ecology, distribution, a summary distribution maps, and a review of areal distribution for each species.

Keywords: lignicolous fungi, mycodiversity, *Amylostereum areolatum*, *A. chaillatii*, *A. laevigatum*, host interaction.

Mycodiversity on Bregalnica River Catchment Area – New Fungi Records for Macedonia

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The Republic of Macedonia is mycological relatively well studied and until now approximately 2,000 macromycetes species are known. Previous systematic mycological researches in the Bregalnica

river catchment area were concerned to the Osogovski Planini, while for several other localities there are only single data in separate papers. A total of 289 fungi species were known for this area.

In the period 2014-2015 mycological field research in various localities and habitats were carried out. Collected species were identified in the Mycological Laboratory (Faculty of Natural Science and Mathematics, Skopje). Morphological analyses were performed based on macrochemical reactions on basidiomes (NH₄OH, KOH, ferric sulfate, etc.), as well as by light microscopy. Taxonomically relevant microscopical characters (spores, basidia, etc.) were visualized in KOH, Melzer's reagent or Congo Red.

According to the previous data, as well as current project researches a total of 629 fungi species in the Bregalnica region are identified. The most reach habitats are beech forests and azonal vegetations, while lower diversity is noted in oak forests, as well as in pine plantings or pine forests. 70 taxa represent a new data for Macedonian mycobiota. The most of them belong to the phylum Basidiomycota (61), while the rest to Ascomycota (5), Zygomycota (1) and Myxomycota (3). According to the substrate 41 species are terricolous, 28 lignicolous and one species (*Spinellus fusiger*) is fumicolous, found on old *Mycena* specimen.

It is important to highlight localities with old and well developed forest associations, such as gorges of the rivers: Trebomirska, Ramna, Pehchevska, Zletovska, Blateshnicka, Gradeshka, Zrnovska Reka and Bregalnica, and the localities Kartal and Pesok, where most of the new fungi records were collected.

Keywords: Macedonian mycobiota, new data, fungal diversity.

Distribution of genus *Hyphoderma* Wallr. (Basidiomycota, Fungi) in Republic of Macedonia

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This paper presents data from systematic research of the genus *Hyphoderma* in the Republic of Macedonia. Species that belong to this genus are decomposers. These fungi cause white rot in wood, which is result of the degradation of cellulose and lignin. During this investigation, 21 species of the genus *Hyphoderma* were registered in the Republic of Macedonia including: *Hyphoderma argillaceum*, *H. cremeoalbum*, *H. definitum*, *H. etrusciae*, *H. guttuliferum*, *H. litschaueri*, *H. macedonicum*, *H. malenconii*, *H. multicystidium*, *H. mutatum*, *H. nemorale*, *H. obtusifforme*, *H. obtusum*, *H. occidentale*, *H. pallidum*, *H. praetermissum*, *H. puberum*, *Hyphoderma radula*, *H. roseocremaeum*, *H. sambuci* and *H. setigerum*. Some of the species are cosmopolites while others are considered to be rare species. These species are found in different forest communities throughout the whole territory of the Republic of Macedonia. Of the total number of species, 7 are newly registered for the mycobiota of our country: *Hyphoderma definitum*, *H. etrusciae*, *H. litschaueri*, *H. malenconii*, *H. multicystidium*, *H. nemorale* and *H. occidentale*. The aim of this research was to gather more complete data regarding the distribution and the ecology of the species within this genus. The list of registered species, including data from relevant literature, information from the collection of fungi MACFUNGI, as well as data field and laboratory research and analyses is presented.

Keywords: *Hyphoderma*, corticioid, white rot, distribution, Macedonia.

Larger fungi of conservation importance from the territory of Rila National Park, Bulgaria

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Data about the distribution of larger fungi of conservation importance in Rila National Park are presented. Rila was declared as a national park during 1999 year. The park is situated in the south-western Bulgaria and encompasses the main parts of Rila Mts on the total area of 81046 ha. Rila National Park is characterized by specific and considerable rich biodiversity, typical for the high mountains. The vegetation cover including the coniferous forests belt, subalpine and alpine belts. The native quasiboreal coniferous forests prevailing (pure and mixed *Picea abies*, *Abies alba*, *Pinus sylvestris* communities) Typical for the Rila Mts are the forests of balcan endemic *Pinus peuce* (95 AO High oro – Mediterranean pine forests, Natura 2000 habitats).

The total number of currently known larger fungi (ascomycetes and basidiomycetes) in Rila National Park is about 700 species. The information on larger fungi was published in 34 scientific papers (22 after 2000). A part of materials was recorded by authors in the framework of Update of Plan for management of the park Project (2015). Sixty five larger fungi are of conservation value, included in the Red List of Fungi in Bulgaria. They are listed in the following threat categories: Critically Endangered (CR) - 31 species, Endangered (EN) - 8 species, Vulnerable (VU) - 17 species, Near Threatened (NT) - 5 species, Data Deficient (DD) - 4 species. Forty species are included also in the Red Data Book of the Republic of Bulgaria. Four species (*Amylocystis lapponica*, *Gomphus clavatus*, *Phylloporus pelletieri*, *Suillus sibiricus*) are threatened at European level and are listed in the Criterion A (ii) species in the Rila IPA in Bulgaria. Three species (*Catathelasma imperiale*, *Ph. pelletieri*, *S. sibiricus*) are listed in the National Biodiversity Act (Appendix 2a). Four species (*Cortinarius violaceus*, *G. clavatus*, *Sparassis crispa*, *S. sibiricus*) are objects of observation in the park in the frame of the National Biodiversity Monitoring System. *S. sibiricus* is characteristic species for the *Pinus peuce* communities in Rila Mts. Two species: *Ditiola radicata* and *Trichoglossum hirsutum* were recorded for the first time in the park by the authors during field studies in the framework of Update of the Plan for management of Rila National Park Project.

Larger fungi of conservation importance have been found mainly in coniferous and beech forests and in peat habitats on the territory of the park.

Keywords: ascomycetes, basidiomycetes, Red List of Fungi in Bulgaria, Rila Mts

Larger fungi in Bistrishko Branishte Biosphere Reserve in Vitosha Mt, Bulgaria

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Data about the species diversity of larger fungi in Bistrishko Branishte Biosphere Reserve are reported. This is one of the first Bulgarian reserves. It was declared during 1934 year and was re-categorized in 1977 as biosphere reserve with a total area of 1061.6 ha. Bistrishko Branishte Biosphere Reserve is situated on the north-eastern slopes of Vitosha Mt, on the territory of Vitosha Natural Park. The vegetation cover including the coniferous forests belt and subalpine belt. Natural acidophilous *Picea abies* forests (Vaccinio-Piceetea – 9410) prevailing. The oldest trees are 140-150 years. There are large windthrow spots in the reserve after the windfall in 2001.

About 215 larger fungi have been recorded so far in Bistrishko Branishte Biosphere Reserve. Of them 30 species belong to *Ascomycota* (3 classes, 4 orders, 13 families, 28 genera) and 185 species belong to *Basidiomycota* (3 classes, 12 orders, 40 families, 93 genera). Order Agaricales dominated by the number of species (100). The information on larger fungi was published in 16 scientific papers (6 after 2000). A part of materials (112 new for the reserve species) was collected by the authors during the field investigations in the reserve within the framework of some projects (2007 – 2015). The greatest fungal diversity was found in pure and mixed spruce forests in the reserve. Wood-inhabiting fungi (saprotrophs and parasites) from the basic phases of the fungal succession on dead coniferous wood prevail on the windthrow spots. *Fomitopsis pinicola* is absolute dominant among all fungal species on live and dead spruce wood regarding the number of fruit bodies and frequency. The presented data were found in the framework of Biomonitoring on windthrow areas and dynamics of biological complex in Bistrishko Branishte Biosphere Reserve Project (2007 – 2011).

Seventeen larger fungi of conservation value, included in the Red List of Fungi in Bulgaria have been recorded in the reserve so far. They are listed in the following threat categories: Endangered (EN) – 7 species, Vulnerable (VU) – 8 species, Near Threatened (NT) and Data Deficient (DD) – 1 species per each category. Nine fungal species are included also in the Red Data Book of the Republic of Bulgaria. Four species: *Amanita porphyria*, *Clavariadelphes ligula*, *Leucocortinarius bulbiger*, *Limacella guttata* were recorded for the first time in the reserve by the authors. Three larger basidiomycetes (*Cortinarius violaceus*, *Gomphus clavatus* and *Sparassis crispa*) are objects of observation in Bistrishko Branishte Biosphere Reserve in the frame of the National Biodiversity Monitoring System. *G. clavatus* is a species threatened at European level and was listed in the Criterion A (ii) species in the Vitosha IPA in Bulgaria.

Keywords: ascomycetes, basidiomycetes, fungal diversity and conservation, Vitosha Natural Park, windthrow areas

***Scleroderma meridionale* (Boletales) in Bulgaria and Greece**

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Scleroderma meridionale is one of the less known European species of the genus, notable for its southern distribution with few collections known from the Balkan Peninsula. In the last few years the author had the chance to study numerous specimens from Bulgaria and Greece and the observations are reported herein. The new collections are described in details and illustrated. Their variability is discussed and the distinguishing characters are outlined. The known worldwide distribution is summarized and briefly discussed. Notes on some aspects of the ecology and phenology of the species are also included. It is suggested that *S. meridionale* is probably more widespread and not uncommon in the southern parts of the Balkan Peninsula, but is under-collected.

Keywords: Balkan mycota, Sclerodermatineae, sequestrate fungi

***Neoboletus xanthopus* (Boletaceae) – a widespread but little known bolete species**

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Neoboletus xanthopus was recently described to accommodate morphologically and genetically distinct collections of the *N. luridiformis* group, to which the variously interpreted name *Boletus discolor* and combinations based on it were applied in the past. Being familiar with this taxon since 2002 and having studied ample collections of this and the related species from Bulgaria, the author shares his observations herein. The variability of *N. xanthopus* is discussed, supplemented with illustrations of representative collections, including scanning electron microphotographs of the basidiospores of the known European species of the genus. The separating characters of *N. xanthopus* are outlined. Peculiarities of the ecology of the species are discussed and compared to those of *N. luridiformis* and *N. pseudosulphureus*. *Neoboletus xanthopus* seems to be widespread and common species, especially in south of Europe, as inferred from numerous Bulgarian collections.

Keywords: Boletales, Boletaceae, *Boletus discoloroides*, termophilous boletes

A contribution to the vascular flora of south Bačka loess terrace (North Serbia)

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Floristic and phytocoenological investigations during last two and a half centuries in Bačka have been focused toward research of specific localities at Subotičko-horgoška sands, Bačka loess plateau, swamps and salines. Because of specific substrates and size of objects itself, areas that have less interesting geomorphological character such are loess terraces, were reduced to sporadic floristic records. There was one comprehensive floristic investigation of northern part of town of Bačka Palanka, during which were registered 168 taxa of the vascular flora. Field floristical investigations during 2014-2016 included the area of south Bačka loess terrace (northeastern and central part of Bačka Palanka Municipality) and area restricted with Dunav-Tisa-Dunav hydrosystem (Bački Petrovac, Maglić). There were recorded 478 taxa of the vascular flora, of which 467 at species level and 11 at subspecies level. Plant taxa were grouped into 96 families and 302 genera. Family with largest number of species were Asteraceae (63), Poaceae (41), Rosaceae (31), Fabaceae (28), Lamiaceae (27) and Brassicaceae (23), while the most numerous genera were *Carex*, *Euphorbia* and *Prunus*, followed by *Potentilla* and *Rumex* and last *Ranunculus* with 8, 7 and 6 species respectively. Obtained areal types spectrum reflected phytogeographical affiliation of investigated area, which belongs to Pannonia province of Southsiberian-pannonian subregion, while the significant contribution of adventive areal type (50 taxa) is consequence of strong antropogenic influence on remnant fragments of natural and seminatural habitats. Biological spectrum is characterized by domination of therophytes and hemicryptophytes. Strictly protected species *Crepis pannonica*, 27 protected plant species and five tertiary relict plants belonging to ecological group of hydrophytes were registered at investigated area. Considering the strong antropogenic influence and supremely devastated habitats, investigated area is characterized with relatively large number of species of vascular flora which encourage further investigations of these floristically “less interesting” and neglected areas.

Keywords: Bačka Palanka, Bački Petrovac, adventive species, *Crepis pannonica*

Epipogium aphyllum - the ghost is back to Serbia again

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Epipogium aphyllum is the only European representative of its genus. The species is an obligate myco-heterotrophic achlorophyllous orchid, that inhabits temperate regions of Eurasia, from the Atlantic to Japan. Although it has a wide range, in almost all regions is present in a small number of localities with populations that often count only a few individuals. Flowering is happening in breaks

that sometimes last for several decades and often under the forest floor, while thanks to the existence of underground stolons plants change their position in the locality. For these reasons it gained almost mystical status in some countries - "the ghost orchid". This species is on CITES List, Habitat Directive, IUCN Red List (LC), while in Serbia it is in the category of critically endangered species due to insufficient data. For the territory of Serbia, the first specimen had recorded by Pančić in 1847 on the Belica Mt near Jagodina, and then on the Rtanj Mt (1859). In addition to these herbarium data, there are also two literature references - Beljanica Mt (1950) and the gorge of the Resava river (1997/98). During the floristic studies of the southwestern Serbia in July 2014, 4 flowering spikes of *E. aphyllum* were recorded on Mokra Gora Mt (Prokletije Mt range), above the village Draga (34TDN454) at 1512 m a.s.l. in the forest of beech, fir and spruce. Registered specimens grew in thick leaf-litter, without other ground vegetation, in a deeply-shaded place right next to the fir tree. If we don't take into account the literature data that have not been yet ratified by the relevant herbarium specimens, this is the first published report of this species in the last 155 years in Serbia.

Keywords: saprophytic orchids, Balkans, Josif Pančić, Mokra Gora Mt

Two New *Stipa* Species Records on Macedonian Ultramafics

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Diversity of the genus *Stipa* on Balkan Peninsula has not been investigated in detail. Reasons for this may lay in the fact that this genus has very complicated infrageneric taxonomy, with the lack of stable differential characters for species delimitation. Also the fact that many different names exist for one taxa makes the situation even more complicated. *Stipa* species distributed on the ultramafic substrates are much more of enigma. The important reason, along with the already mentioned ones, lies in the fact that these rocks are very discrete units in terms of their distribution, so the taxa thriving on them are somehow isolated. Two *Stipa* species new for Republic of Macedonia are found on ultramafics in northern part of the country, between Veles and Katlanovo. *Stipa crassiculmis* subsp. *picientina* has been known as endemic for southern Italy and Sicily, but was found on ultramafics of Macedonia last year. This is the first record of this taxa on Balkan Peninsula, so it should be considered as Balkan-Appennine subendemic species. Species *Stipa ucrainica* was also recorded nearby. This taxon is distributed in Eastern Europe and Asia Minor. Recently, the species was recorded in Bulgaria, in 2008. Both taxa form stands in which they appear as dominant species. These stands are very xeric and with stepic character.

Keywords: *Stipa crassiculmis* subsp. *picientina*, *S. ucrainica*, diversity, ultramafic rocks, steppe, relics

***Sesleria ujhelyii* Strgar (Poaceae), a neglected species of the Balkan Peninsula**

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Sesleria ujhelyii is a neglected taxon, described by Strgar (1973) from the material collected in Bosnia and Herzegovina, site Dariva on Miljacka River. It belongs to taxonomically interesting *Sesleria juncifolia* species complex, an endemic Balkan-Apenine group. Despite of clear differences in leaf anatomy with respect to other taxa belonging to this complex, elaborated by Strgar, relevant databases and plant checklists treat this species either as a synonym of *S. juncifolia*, or it has been completely neglected. The aim of this study was to describe the variability of morphological and anatomical characteristics of populations of *S. ujhelyii* from the Balkan Peninsula, with comparison to other populations within the complex *S. juncifolia*.

Our study has shown that the most significant characteristic for distinguishing *S. ujhelyii* (among other species in the complex) is the presence of the continuous sclerenchyma on abaxial side of the tiller leaf, supporting the studies carried out by Strgar. The presence of the continuous sclerenchyma on abaxial side was confirmed in the populations in Western Bosnia and Herzegovina (population from locus classicus), in eastern part of Serbia and in the South of Macedonia (population from Galičica Mountain). Populations of *S. ujhelyii* differed slightly in respect of various morphometric characters. The characteristics of leaf anatomy, as in case of many other grasses, have proven to be crucial for distinguishing *S. ujhelyii* from other species in the complex.

Keywords: Balkan Peninsula, continuous sclerenchyma, leaf anatomy, Poaceae, *Sesleria ujhelyii*, taxonomy

Species diversity and conservation significance of *Asteraceae* in the mountain regions along the Bulgarian-Macedonian border

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Asteraceae is one of the largest plant families worldwide. Also, it is the richest in species taxonomic group both in the Bulgarian and Macedonian floras. The aim of the present study was to reveal the taxonomic diversity in the family in the mountain regions along the Bulgarian-Macedonian frontier

as well as to locate the occurrences and assess the population state of the taxa of conservation concern. Field trips were carried out in the following mountains – Osogovo, Vlahina, Malashevka, Ograzhden and Belasitsa in the period 2014-2016. As a result, more than 200 species have been recorded. The share of taxa of conservation concern is relatively small – ca. 2%, however, the area is important for the conservation of these species, e.g. of *Achillea chrysocoma*, *Cota macranta* and *Jacobaea subalpina*, since it shelters the only or some of the largest populations of these taxa in Bulgaria and Macedonia. The poster presents analysis of the taxonomic diversity in the family, endemism, as well as provides data about the populations and the threats to the species of conservation concern.

Financial support under the project ‘Floristic and taxonomic studies of selected genera from the families Lamiaceae and Asteraceae in the border regions of Macedonia and Bulgaria’ is gratefully acknowledged.

Keywords: Bulgarian flora, Compositae, Macedonian flora, plant conservation, vascular plants

Impact of the alien species *Fallopia × Bohemica* (Polygonaceae) on the native plant diversity in Bulgaria

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Fallopia ×bohemica (Polygonaceae) was introduced to Bulgaria for use as an ornamental plant in urban environments. However, it escaped from cultivation and spread to numerous localities across the country growing in different habitats – mainly in urban areas and on riversides, occasionally on roadsides and along railways, usually in sunny spots but also it withstands partial shading. The species has been recorded in 8 out of 20 floristic regions of Bulgaria – Black Sea Coast (*Northern*), Danubian Plain, Forebalkan (*Western*), Balkan Range (*Western, Central*), Sofia region, Znepole region, Vitosha region, Rhodopi Mts (*Central*). The aim of this study was to reveal the impact of this invasive alien on the native flora and vegetation. For this purpose 1×1 m plots with and without the invader were compared; in each locality 10 pairs of plots were studied. The following parameters were recorded: total number of vascular plant species in each plot, total vegetation cover, cover of each plant species, and maximal height of each species. The results suggest *Fallopia ×bohemica* is a successful competitor, significantly decreasing the available space to local plants, changing the composition and structure of the plant communities, and reducing the floristic diversity in the invaded areas.

Financial support by the Financial Mechanism of the European Economic Area (2009–2014), Programme BG03 ‘Biodiversity and Ecosystem Services’, under project IBBIS (Contract Д-33-72/20.07.2015) is gratefully acknowledged.

Keywords: Bulgarian flora, ecological impact of alien plants, invasive species, Reynoutria

Orchid flora of the Natural Monument “Sopotnica Waterfalls” (Southwest Serbia)

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Natural Monument “Sopotnica Waterfalls” is located in Southwest Serbia, on the western slopes of the Jadovnik Mt, in the municipality of Prijepolje. The protected area covers approximately 2 km² along the river Sopotnica. Field researches were conducted in all growing seasons from 2014 to 2016 year, during which 28 representatives of the family Orchidaceae were registered. This puts family Orchidaceae to the second place by the plant species richness in the investigated area. There are representatives of 15 genera (Serbia 22), wherein *Orchis* (5) and *Epipactis* (4) are the species-richest genera, followed by six genera with two species and seven with one species. The analysis that included number of orchid species in relation to the surface area and total plant species number, pointed out a high orchid diversity, which makes Natural Monument “Sopotnica Waterfalls” one of the centers of diversity of this family in Serbia. All species are geophytes, among them three are obligate saprophytic plants (*Neottia nidus-avis*, *Limodorum abortivum* i *Corallorhiza trifida*). Species of Centraleuropean (8) and Eurasian (9) floral elements inhabit various types of habitats, while the Mediterranean-submediterranean (6) and Subatlantic-submediterranean (4) are limited to open habitats (rocky terrain, meadows, scrubs). The only boreal species (*Corallorhiza trifida*) occurred exclusively in the beech forests. All registered taxa, except three species - *Ophrys insectifera*, *Epipactis purpurata* and *E. leptochila*, are protected on the territory of Serbia. Populations of most registered species counted a hundred or even more of flowering individuals; this, together with good protection measures that are implemented in the field and preserving of the traditional way of life of people in this area ensures the survival of these globally threatened species in the area NM “Sopotnica Waterfalls”.

Keywords: Prijepolje, diversity, *Orchis*, *Epipactis*

Phytogeography characteristics of orchids (Orchidaceae) in Croatia

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The Orchid family embraces around 181 taxa in Croatian flora. The goals of this research were to assess the spatial distribution and species richness of Croatian orchids and to predict if the changes in distribution are dependent to climatic changes. These goals were achieved by Species Distribution Models and combining them with botanical collection records and ecological variables using presence – only data algorithm, MAXENT. The results indicate higher species richness in coastal parts (Mediterranean and Alpine biogeographic regions) and lower species richness in eastern continental parts.

Keywords: Maxent, Species distribution modelling, orchids

***Osmunda regalis* (Royal fern) in Bulgaria – population status, threats, and conservation**

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Osmunda regalis is among the rarest vascular plants in the Bulgarian flora. It is legally protected by the Bulgarian Biodiversity Act and included in the national Red Data Book and Red List as “Critically Endangered”. The aim of our study was to establish the current state of *O. regalis* in the Bulgarian flora and to contribute to the conservation of the species and its habitats.

Information on the ecological requirements, population and habitat characteristics and state, and plant communities was collected *in situ*. Monitoring was carried out. Chromosome number was counted in root tip squashes stained with haematoxylin.

Royal fern has extremely restricted distribution in Bulgaria, in the Valley of River Struma floristic region. So far only 3 localities have been known. A new one was found during the investigation. Most locations are very closely situated and it is likely they are remnants of a larger strongly fragmented population.

Diploid chromosome number of $2n=44$ is reported for the first time from a Bulgarian accession.

Habitat requirements are specific – open or shady places, marshy grounds, along streams and canals, wet places in black alder woods. The populations occupy small areas, comprising 30–250 plants growing mainly in groups.

Some of the most important limiting factors or threats for the species are: loss and degradation of its habitats due to anthropogenic and natural drainage; successional changes leading to overgrowth by competing species (mainly *Pteridium aquilinum*); limited range in Bulgaria; low reproductive, dispersal and competitive potential of the species.

The investigation showed that urgent conservation measures must be undertaken. Royal fern and its habitats need *in situ* restoration and maintenance of a suitable water regime, and control of competing species. Search of new localities and long-term monitoring of the known ones are needed.

Keywords: Bulgarian flora, distribution, endangered species, monitoring

Floristic diversity of Ovčar-Kablar Gorge (Western Serbia) – last six years of research

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Ovčar-Kablar Gorge is mountainous area which is part of the young fold mountains of Dinaric system; it is located in the western Serbia, between the cities of Čačak and Požega. This region has been placed under official protection as a Landscape of Outstanding Features „Ovčar-Kablar Gorge“ in the

year of 2000 and covers the area of 2250 ha. A constant negative human influence in this region is mostly reflected on forest ecosystems, in mind of sustainable management and use, on meadows and pastures which are characterized by conspicuous succession and river ecosystems endangered by construction of weekend settlements. The previous floristic research of LOF „Ovčar-Kablar Gorge“ were carried out periodically and partially with large breaks even for several years, so the aim of our study was to record and add new data to existing floristic list of these vulnerable ecosystems, during the period from 2011-2016. Results showed the presence of 345 taxa at species level and 12 taxa at subspecies level. Species richest genera were *Trifolium*, *Campanula*, *Veronica*, *Lathyrus* and *Asplenium*, while the most species abundant families were Fabaceae, Lamiaceae, Rosaceae, Asteraceae and Poaceae. The biological spectrum was characterized by the dominance of hemicryptophytes, while phytogeographical analysis showed the significant share of Central European, Eurasian and Mediterranean-Submediterranean areal type. Six strictly protected, 45 protected and nine relict species were registered as well as some invasive species such as *Ambrosia artemisiifolia* and *Reynoutria sp.* Due to the presence of great natural wealth, preserved flora and occurrence of some alien and invasive species, further floristic researches should be directed primarily to the recording of alien species and sensitive habitats.

Keywords: vascular flora, anthropogenic influence, tertiary relicts, invasive species

***Galanthus elweisii* Hook, a new species to the flora of Kosovo**

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Flora of the southern part of Kosovo is studied in a limited way due to the difficult terrain and the fact that up to 90th it was a military area. In this article, *Galanthus elweisii* is registered for the first time in Kosovo. This species is assembled in Restelica, on the road leading to the border with Macedonia, or between this street and the Albanian border with Shishtavec. This species is found in different habitats mainly siliceous substrate or wet meadows of Dragash. This territory belongs to phytogeographic system Skardon - Pindik (Shari-Pindi) and includes the mountain ranges of Sharri (Kosovo Vraça Mountains-Malet Vraça të Kosovës) and Korabi in the study area. Presence, description, spread and mapping of this endemic and so far unreported species for the territory of Kosovo, is reviewed and discussed in this paper.

Keywords: new species, spread, Kosovo, Macedonia, Albania

Does differentiation between *Typha* species in terms of micromorphological characters exist?

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In order to determine a degree of morphological differentiation of three *Typha* species, *Typha latifolia* L., *Typha angustifolia* L. and *Typha laxmannii* Lepechin, micromorphological characters of foliar epidermal tissue (number, length, width and surface of stomata, surface of epidermal cells) were studied. The plant material was collected on native habitats in the area of southeastern Serbia. Statistical techniques including descriptive statistic, ANOVA, PCA, CDA and clustering UPGMA based on Mahalanobis distances were performed. The lowest coefficient of variation is shown by the following characters: length and width of stomata. PCA and CDA indicated slight differentiation of *Typha laxmannii* population in comparison with other populations. The only character that significantly contributes to differentiation along the first PCA axis is surface of stomata on abaxial side of lamina. The results of statistical analyses have shown that studied species have low degree of differentiation based on micromorphological characters.

Keywords: cattails, morphological separation, epidermal tissues, central Balkan Peninsula.

Distribution range of the plant species *Pistacia terebinthus* in the Sharr Mountain

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This summary gives data about a location of the plant species *Pistacia terebinthus* on Sharr Mountain. *Pistacia terebinthus*, is plant species which by local researchers is found only in one location in the Shar Mountain (Jazhincë). In our scientific research conducted during the vegetation period (March 2013, 2014, 2015, 2016), this rare plant species is also found in another location in the Shar Mountain (near the village of Novo Selo). The locality in which *Pistacia terebinthus* was found is important for conservation ecology because even after the opening of the mountain road and near this street was built a channel for delivery of water to the artificial lake Mavrovo this plant species could survives even today in the locality mentioned above.

Keywords: Plant species, *Pistacia terebinthus*, location, Sharr Mountain.

An insight into Mediterranean component of the Serbian bryophyte flora

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Serbian bryophyte species diversity has significantly increased during the last decades, with few dozen new species records for the territory of Serbia. However, biogeographical approach to the bryophyte flora has been neglected and the chorological spectrum of Serbian bryoflora is unclear. Though Serbia belongs to peri-Mediterranean region, due to highly diverse orography it has mainly temperate habitats, and significantly less Mediterranean-like ones. The distribution type analysis of the known Serbian bryoflora showed that more than 11% of the total bryophyte species belong to Mediterranean ones in wider sense (incl. submediterranean distribution types). However, strict Mediterranean species (two liverworts and 5 mosses) represent 1.02 % of Serbian bryophyte flora and thus deserve special conservation attention, as well. In wider sense, 6.3% of Serbian liverworts and 12.2% of moss species belongs to (sub) mediterranean distribution type(s). Among these, number of rare and conservationally important species is considerably high; 12 species are internationally important and included in the Red data book of European bryophytes. Rather more than those 12 species are candidate for the New National Red List, but further investigation and records, national distribution, ecology and biology is needed.

Keywords: mosses, liverworts, chorological types, Mediterranean, conservation, diversity

Comparative analysis of epilithic diatom assemblages of springs and streams in the Konjuh Mountain (Bosnia and Herzegovina)

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Springs are recognized as hotspots for freshwater biodiversity conservation. The objectives of the study were to analyze and compare the biodiversity of epilithic diatom communities of spring and stream habitats in the Konjuh Mountain. The measuring of basic physical-chemical parameters and diatom sampling were carried out in the summer and autumn in 2013. We sampled diatoms in small mountain springs (5 localities) that flow into headwater streams (other 5 localities) by scrubbing the rocks using standard methodology. Relative abundances of diatoms were established by counting up to 400 frustules on permanent slides prepared in Naphrax.

Spring habitats were more diverse in diatoms ($H' \log_e = 1.67$ average) in comparison with headwater stream habitats ($H' \log_e = 1.15$ average), which was confirmed by *t*-test ($t=2.79$; $p=0.013$). The similarities of community structure from different habitats were investigated by non-metric

multidimensional scaling, on the Bray-Curtis similarity matrix in software package Primer 6. Using Permutational Manova test, it was proved that there is a difference in the structure between spring and stream communities (Pseudo $F=5.39$, $p=0.039$), while the difference in the structure of communities in the summer and autumn statistically was not confirmed ($F=1.819$, $p=0.222$). The most abundant and frequent species in spring habitats were: *Achnanthydium minutissimum*, *Cocconeis pseudolineata*, *Gomphonema pumilum* var. *elegans*, *Meridion circulare* and *Planothidium lanceolatum*. The most frequent species in headwater stream habitats were: *Achnanthydium minutissimum*, *Cocconeis placentula* var. *euglypta*, *Cocconeis placentula* var. *lineata* (and *Cymbella tridentina*).

The springs are heterogeneous habitats with specific flora of diatoms due to more stable waterflow in comparison with streams. Although the springs cover very small areas, the diversity of species that inhabit springs must not be neglected in planning the use of these natural resources.

Keywords: diatoms, community, epilithon, spring, stream, diversity

European Roller (*Coracias garrulus*) prey on breeding territories in Vojvodina (North Serbia)

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Breeding site selection and reproductive success of obligate secondary cavity breeding bird species, European Roller (*Coracias garrulus*), depends mainly on presence of cavities for nesting and prey availability. The European Roller population is declining throughout Europe, and such trend almost led to local extinction in North Serbia in second half of 20th century. During last 15 years, however, Serbian population is successfully recovering due to installing large number of nest boxes within suitable habitats. Understanding how habitat quality, including prey availability, influence the site selection and reproductive success of European Roller is crucial for planning of future conservation action. We surveyed arthropod communities (available prey) around occupied and unoccupied nest boxes, and on potential territories located in still unoccupied suitable landscapes without nest boxes. Also, we collected and analyzed prey remains found in occupied nest boxes (used prey). Survey is conducted in the mosaic farmland landscape in North Serbia, dominated by grasslands. Occupied and unoccupied nest boxes were located in central Banat area, while potential territories were in south-east Bačka and Srem area. Differences in prey community and its abundance between occupied, unoccupied and potential breeding territories were tested. Our results have shown no significant difference in available prey community and its abundance between these categories. Dominant groups in samples of both available and used prey were insects of order Coleoptera and Orthoptera. These results indicate that in North Serbia available prey was not key factor in Roller selection of a nest box, and it is not limiting factor for further expansion to chosen suitable sites outside of current breeding range. Installing more nest boxes in unoccupied suitable sites in North Serbia is suggested conservation strategy, since our results reveals pattern of habitats with sustainable amount of available prey lacking in nest cavities, the key limiting factor for the species.

Keywords: *Coracias garrulus*, nest boxes, arthropod communities, breeding site selection.

Monitoring of Vertebrate Fauna and Conservation Problems in Parts of Sofia Plain - Preliminary Results

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Although Sofia plain is heavily impacted by human activities, surrounding the largest city in Bulgaria, the territory holds a large diversity of animals and plants, including many conservation dependent, rare and endangered species. At the same time, the potentially dynamic changes in the species composition and conservation threats in the area are not constantly studied in the recent years.

The following research presents results from six-month monitoring of the vertebrate fauna and biodiversity-threatening factors in the Eastern part of Sofia plain, conducted by student's club Skorec of Sofia University and colleague of New Bulgarian University.

During the research transect of 55 km was covered, which includes a variety of habitats: settlements, agricultural areas, deciduous forests, wetlands. Full data was collected including species composition and their biological and ecological characteristics. Conservation problems were also identified. The information was analyzed by statistical software SPSS v.21, SigmaStat 3.5 and SigmaPlot 11.0.

As a result of our work we identified 8 species of fishes, 3 species of amphibians, 2 species of reptiles, 94 species of birds and 10 species of mammals. Some of the birds were confirmed for the first time in the area (*Sterna albifrons*), or were observed rarely in Sofia plain in general (*Anthus cervinus*). Additionally, conservation threats like disturbance by vessels and fisherman in Ognyanovo dam and animal road kills were identified and constantly monitored. Most of the mammals identified in the area were found dead on roads (*Martes foina*, *Meles meles*, *Canis aureus*, *Erinaceus concolor*, *Apodemus flavicollis*, *Rattus norvegicus*).

Keywords: ecology, zoology, tetrapods

Contribution to the knowledge of the Lycaenidae fauna of Central part of Kosovo

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Kosovo has a rich and diverse fauna of butterflies which, recently, is arising a huge interest of biodiversity researchers. In this paper, the results of the research of butterflies of the Lycaenidae family, conducted in the mountains of Ceri, Stermica and Kryeguri, in the central part of Kosovo, are presented.

In total twenty-five species of Lycaenidae have been identified. Butterflies were collected in the period May – July 2016 in 11 localities. All the recorded species are listed in the red list of IUCN,

out of them 23 belong to the LC category, *Phengaris arion* (Linnaeus, 1758) to EN (Endangered) and *Polyommatus eros* (Ochsenheimer, 1808) to the NT (NearThreatened).

Keywords: Lycaenidae, Kosovo, threatened species

Endangered species of the Zapadna Morava River ichthyofauna (Central Serbia)

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The hydrographic network of the Republic of Serbia is quite diverse, with 66,000 km of rivers and 150 reservoirs. Highly variable hydroecological conditions have led to high ichthyofaunal diversity. The presence of 95 fish species of 27 families has been registered. The Zapadna Morava (298km) is a central component of the Serbian hydrosystem. The river flows through the central part of the country characterised by intensive urban, industrial and agricultural activities which cause water pollution and other negative impacts. These factors have an adverse effect on hydrobiont communities, including fish assemblages. Two reservoirs constructed on the river (Ovčar Banja and Međuvršje) have undergone severe changes, primarily related to sedimentation and intensive eutrophication. Periodic ichthyological studies were conducted during 2010-2014 in the upper and middle courses of the Zapadna Morava River. Fish specimens were caught using standard fishing nets with different mesh sizes and electrofishing equipment. During the period, the ichthyofauna of the Zapadna Morava River included 28 species of 9 families. The family Cyprinidae predominated in terms of the number of registered species (18) and number of specimens caught (93.28% total catch). Bleak (*Alburnus alburnus*) was a dominant member in the fish assemblage, followed by chub (*Squalius cephalus*), roach (*Rutilus rutilus*), common nase (*Chondrostoma nasus*) and Prussian carp (*Carassius gibelio*). Some native species, primarily zingel (*Zingel zingel*) and tench (*Tinca tinca*), are facing extinction. These species are extremely threatened due to water quality deterioration, dam construction, river bed regulation, high water fluctuations, sediment deposition and other factors. A total ban on zingel and tench fishing was introduced in Serbia in 2009.

Keywords: fish assemblage, zingel, tench, protection

Additional morphological data of *Gammarus orientalis* S. Karaman, 1934 from type locality, Derbend, Erdschias Dag, Asia Minor

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The species *Gammarus orientalis* was described briefly under the name *Gammarus balcanicus orientalis*, ssp. n. by S. Karaman (1934) from torrent on 2100 m. above sea level in Derbent, on E. slope of Erdschias-Dagh Mt., E. Asia Minor. The numerous taxonomic characters of the male of this species are not published, and its female was not described. Later, this taxon was considered by many authors a synonym of *Gammarus balcanicus* Schäferna, 1922 [loc. typ. Kolašin, Crna Gora], together with many other known species and subspecies similar to *G. balcanicus* (G. Karaman, 1977H; Karaman & Pinkster, 1977; etc.). Recent studies of various scientists [Mamos, Grabowski, Wattier 2015, etc.] indicated that *Gammarus balcanicus* (sensu auctorum) represent complex of numerous different taxa similar to each other but genetically different. In this light, it is important to recognize taxonomic characters of known members of *G. balcanicus* Complex of taxa. We redescribe *G. orientalis* based of typical material.

Keywords: Amphipoda, Gammarus, description, Asia Minor, taxonomical characters

New record of *Umbra krameri* Walbaum, 1792 in Serbia

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The European mudminnow (*Umbra krameri*) occurs in the lowlands of the Danube River drainage, from Vienna, Austria to the Black Sea, and in the lower reaches of the Dniester drainage (Ukraine and Moldova). It is on the Red List of many European countries and it also appears in Appendix II of Bern Convention and in Annex II of Habitats Directive. Drainage of wetlands to reclaim arable land has posed a great threat for this species. Two locations in Serbia where European mudminnow is currently reported are Kupusinački Dunavac, near Sombor, and Zasavica, near Sremska Mitrovica. Special Reserve „Kraljevac“ is located in the southern part of Banat, on the edge of Deliblato sands. Water reservoir Kraljevac was formed in the beginning of 1980s when the original fen habitat in alluvium near Deliblato was dammed. This lake with surface area of 162ha and average depth of 2,5m irreversibly changed original habitat, leaving only a small part of the original fen habitat in the north-western part of the reserve, where the fish sampling was conducted for the purpose of this study. Coordinates of the sampling site are N 44°51'06.9" E 020°58'58.1". In total, 28 specimens of *Umbra krameri* were sampled, aging between 0+ and 4+ with average TL 71.49 mm (1SD: 13.88 mm), average SL 59.18 mm (1SD: 12.14 mm) and average mass 4.18 g (1SD: 2.90g). European mudminnow was accompanied by: pike, roach, rudd, perch and sun bleak. We conclude that it is necessary to perform further population

size and distribution analyses. In addition, our sampling site was on north-western border of Reserve and, in close proximity, we observed similar habitats which are not included in the Reserve but could be inhabited by *U.krameri*. If later would be proven, it could bring to expansion of Special Reserve “Kraljevac”.

Keywords: Umbridae, Pannonian plain, Danube, Kraljevac, endemic species

Ostracoda (Crustacea) in lotic mountain-river habitats in Serbia

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Small crustaceans belonging to class Ostracoda are present in various aquatic habitats worldwide. Freshwater ostracods have calcified bivalved carapace, and they inhabit ponds and lakes mostly as nectobenthos and crawlers. In fast-flowing rivers and streams, their presence is limited to sheltered places with calmer water and groundwater habitats, such as interstitial waters connected to riverbeds. With the aim to explore their presence and ecology in highland rivers in the mountain region in Serbia, special attention was devoted to their separation from the samples collected in the study of benthic communities in mountain rivers during 2011 – 2012. Invertebrates are collected with Surber-net, taking three samples per locality. Basic physical and chemical parameters are measured on site, and water samples are also collected for further laboratory analyses. To relate occurrence of detected species with measured environmental parameters, Canonical Correspondence Analysis was performed with MVSP by Kovach Computing Services. Ostracods are found in samples from Radovanska, Crnica, Vrla and Mlava rivers, where 5 species belonging to 3 genera are identified. The most frequently sampled species were *Candona candida* and *Psychrodromus olivaceus*. *Candona neglecta* was found in two samples, while *Psychrodromus fontinalis* and *Prionocypris zenkery* were recorded only once on different sites. CCA revealed that the ostracod assemblages were mainly affected by measured conductivity. The new findings of named species present significant contribution to the knowledge on diversity of ostracod fauna in the central part of the Balkan Peninsula.

Keywords: ostracod assemblages, *Candona*, *Prionocypris*, *Psychrodromus*, water parameters

Floristic diversity of sand dunes of Serbia

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Analysis of floral diversity of sand dunes of Republic of Serbia was based on data collected in the field during the period from 2011 to 2016 and available literature data. The analysis included: Subotica-Horgos, Deliblato sands and sand dunes on the right bank of the Danube, near the Iron Gates Gorge - Kladovska, Požeženska and Ramsko-Zatonjska.

Life forms were determined by Ranunkiev (Raunkiaer, 1934).

Floral elements are defined by Gajic (Gajic, 1980), and in exceptional cases according Soó (Soó, 1968).

Representation of floristic genera on deposits of sand in our country is relatively uniform character. As might be expected, generally the most common representatives are annual and perennial grass genera *Festuca* L. and *Carex* L., which are the basic edificators plant communities on the sand. From this floristic image, Subotica-Horgos sands distinguishes partly, because there are the most common genera *Silene* L. and *Alyssum* L. which also contain very important representatives of sandstone flora, mainly in pioneer communities. At the Deliblato sands and sands of eastern Serbia are numerous species of the genus *Trifolium* L., especially present in the various successive stages of successive and regressive vegetation types.

The representation of life forms on the sands in Serbia is relatively uniform. At the same time, the Subotica-Horgos and Deliblato sands are characterized by almost identical biological spectra- most common are hemicryptophytes, a little less represented therophytes, while on the sands of eastern Serbia hemicryptophytes and therophytes almost equally represented. Chamaephytic, geophytes, bryohamephites, phanerophytes, nanophanerophyte and lichens on all analyzed sands are fewer life forms, but a large percentage of its cover, retain a significant role in all plant communities.

All sands in Serbia contain Pontic floral elements in greatest percentage. They are followed by Eurasian flora elements, while Centraleuropean take the third place. Representation of Balkan and Eastern sub-Mediterranean elements varies according to the geographical location of the analyzed habitats and they are the most common in the sands of eastern Serbia.

Sandstone habitats are under constant degradation. This is particularly the case in the sands of eastern Serbia, since they are not under the protection regime. Here are the most common initial forms of vegetation - the pioneering and regressive.

Keywords: sand dunes, floral diversity

Bryophyte diversity of Bulgaria – 40 years after the first Flora

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The first bryophyte Flora and the first comprehensive summary of bryophyte diversity of Bulgaria was made by Sl. Petrov in 1975. This keystone work treated 670 species.

Our aim is to make an overview of the studies of bryophytes in Bulgaria in the past 40 years with emphasis on bryophyte diversity and conservation efforts.

In the last 40 years 118 species were added to Bulgarian bryophyte flora mostly due to finding of new species and to a lesser extent due to taxonomic changes. At present, the total bryophyte diversity in Bulgaria comprises 788 species (193 hepatics and 595 mosses). Of them 251 species are red-listed at the national level. 148 species are rare at the European level and are candidates for the new European Red Book. A map of the spatial distribution of newly found species, as well as reports before 1975 is presented.

The most important conservation measures specifically directed towards bryophyte conservation in Bulgaria are inclusion of 6 species and genus *Sphagnum* in Bulgarian Biodiversity Act, preparation of Action plans and designation of protected areas for conservation of particular species and their habitats, long-term monitoring of selected threatened species included in National Biodiversity Monitoring System.

We also present some of the most recently found species with data on their distribution and ecology in Bulgaria.

Keywords: bryophytes, bryophyte conservation, hepatics, mosses, new findings

Contribution in determination of the methodology for choice of location and dimension of bio corridors for animal across infrastructures transport objects

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Republic of Macedonia is tending to improve state road network with construction of new highways and expressways with main aims better connectivity and safer roads. Frequently asked question is: How to design/ensure animal crossing over the new constructed road? Search out and determinate methods to be use in order to ensure safe crossing of animals. Application of analytical methods in early stage during designing projects in order to ensure safer animal crossing over new constructed roads is a good way to solve this problem.

Macedonia has no experience in applying mitigation measures for animal protection, has no data for animal mortality, still designed and proposed mitigation measures have not been implemented and most important awareness for this issue is not on a intended level to all stakeholders involved in road

projects. This paper aims to contribute in a finding out methodology and determination of a places and types of objects should be apply during designing and building objects for different species that are in danger with the realization on road project construction.

Keywords: biodiversity, fragmentation, construction, green bridge, roads

Identified threats and conservation measures for the priority habitat type *9562 Grecian Juniper Woods in Prespa National Park, Greece

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According to the EU Habitats' Directive (92/43/EEC), habitat type 9562 *Grecian Juniper Woods (GJWs) has been classified as priority and is found only within the Prespa National Park in Greece. Threats for this habitat type have been identified as a result of long-term reduction and/or abandonment of traditional agroforestry practices. Major threats include (a) the encroachment of Grecian Juniper Woods by broadleaved species, (b) an increased fire risk, as a result of increased dead organic material and accumulation of solid waste and (c) low regeneration of juniper species.

Restoration and conservation measures are currently applied under a LIFE funded project (LIFE12 NAT/GR/539 - JunEx) since 2013 and the main actions of the project concentrate on (a) the progressive application of appropriate silvicultural treatments in order to remove broadleaved vegetation, (b) measures to assist the re-introduction of grazing for containing the re-growth of broadleaved vegetation, (c) removal of dry biomass and solid waste from the Grecian Juniper Woods and (d) measures that will enhance the regeneration of juniper species. Additionally, the results of the monitoring actions and the outcomes of the project may be incorporated into management plans, which are formulated and applied by relevant authorities, ensuring the long-term conservation of the habitat.

The GJWs are found within the protected areas of all three littoral countries of the transboundary Prespa basin (shared by Greece, Albania, FYR of Macedonia), and the experience gained can be transferred to relevant stakeholders, if the application of similar conservation measures is rendered necessary in the future.

Keywords: priority habitat type, Grecian juniper woods, agroforestry, conservation measures

Peatland vegetation succession enhances isopod biodiversity

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Peatlands in Croatia are small in size and isolated habitats present on the southern boundaries of their distribution. Vegetation succession, abandonment of traditional land-use and climate change cause their endangerment and loss in the Western Balkans. Specific soil conditions present on these habitats (high moisture, low nutrients, and pH extremes) give an extreme environment for inhabitation to rare but adapted species, both plant and animal, emphasizing specific soil invertebrates, including isopods. Our aim was to explore whether, and to what extent, environmental factors (soil moisture, temperature and pH) and consequently plant associations promote isopod biodiversity. Plants and terrestrial isopods (Isopoda: Oniscidea) were studied in the alkaline fen and adjacent habitats undergoing vegetation succession, located in west-central Croatia. Field work was carried out during 2008 and 2009. Vegetation was sampled using standard European Braun-Blanquet method (extended by Barkman) while isopods were collected using pitfall traps. Vegetation analysis showed three different associations, one for the fen, two for successional stages and one for the beech forest. In fen vegetation high plant species richness was noted. It increased towards the successional stages and decreased in the beech forest. Similar pattern was observed with the isopod species richness and activity density. Six species were found for the researched area, but only five were present on the fen and in the beech forest. Total number of individuals was 1069, out of which the highest values were noted on sites undergone by vegetation succession, while on the fen and in the beech forest the values were considerably smaller. The recorded plant and isopod assemblages show the significance of specific environment present on peatland habitats and the importance of supporting long term efforts to preserve these and surrounding habitats for biodiversity enhancement.

Keywords: Peatland vegetations, isopods, diversity

Mali Ston Bay – a perfect example how to destroy the protected area?

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Mali Ston Bay is a coastal region in the southern Croatia (eastern Adriatic) where the European flat oysters (*Ostrea edulis*) and mussels have been cultivated for centuries. In 1983, due to its ecological and economical importance, the area was proclaimed a special reserve, both in marine and terrestrial zone. The European flat oyster has high economic potential in the European Union market because it has almost disappeared over a European territory due to outbreaks in farms caused by bonamiosis and markeitiosis. Mali Ston Bay is a rare remaining habitat of European flat oysters in Europe and it has

optimal conditions for the reproduction and production of shellfish in relation to the highest standards and health requirements. In 2016, Dubrovnik-Neretva County and Municipality of Ston have accepted of amendments to the Spatial Plan for the construction of tourist resorts in the Bay on the surface area of 13 hectares with the capacity of 550 tourist beds. The Croatian *scientific community* strongly opposed the proposals and has advised to invest in tourism infrastructure on the southern slopes of the Pelješac Peninsula, where the coast for this type of activity is more attractive and thus encourage the development of two complementary economic activities which in principle cannot successfully coexist together.

Keywords: natural heritage, European flat oyster, tourism development, Pelješac Peninsula, south Croatia.

Habitat Monitoring Program in the Area of Tara National Park in 2016

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National Park Tara was declared in 1981 with the total area of 24.991,82 ha. The wider area of Tara has been identified as IPA, IBA, PBA, a pilot area for NATURA 2000 in Serbia, as well as a significant area within the EMERALD network. Woodland, forest and other wooded land habitats are thoroughly explored previously, since Tara is an exclusive forest area in Europe. During July 2016, in cooperation with Management of the National Park Tara and Serbian Ministry of Agriculture and Environmental Protection, we developed monitoring program for mires, bogs and fens, grasslands and lands dominated by forbs, mosses or lichens and inland unvegetated or sparsely vegetated habitats on plots 5 x 5 m, on 75 sites. Habitats were classified using Serbian national classification system. Distribution maps of investigated habitats were made. Dominant and characteristic taxa vouchers were deposited in Herbarium BEOU. Obtained results should give better insight in the state of open habitats within the park and serve as a basis for conservation measures and development of the Park, as well as cooperation with other agencies and the public, for the long-term protection of park ecosystems. Long term monitoring results, together with an effective education program, could improve the quality of life issues affecting not only Park, but also surrounding communities, resulting in significant improvement of the environmental health of the nation.

Keywords: nature, management, habitat classification, protected areas, open habitats

Monitoring of species of conservation interest in the area of Tara National Park (Serbia) in 2016

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National parks represent natural laboratories for comparison of the data with disturbed areas. Tara National park (western Serbia), with about 1000 vascular plant species recorded, almost one third of the total Serbian flora, represents a massif refuge with a great number of Balkan endemic and relict species. Some species are present in Serbia only on Mt. Tara, e.g. *Cardamine serbica*, which population is very endangered and restricted to a small area in Perućac locality. In July 2016. we have mapped over 50 endangered, endemic or relict vascular plant and bryophyte species within the Park area (mainly in open habitats) and collected data on population size, risk factors and their habitats. The project was supported by National Park Tara and Ministry of Agriculture and Environmental Protection. Distribution maps of all investigated species were produced and voucher specimens were deposited in Herbarium BEOU. Detailed distribution maps and recorded population size data should serve for development of the scientifically sound information about the current status of the important plant species within the Tara National Park in order to harmonize nature conservation and sustainable development and progress of the Park.

Keywords: endangered, endemic and relict species, protected areas

Species distribution modelling as a tool to estimate effectiveness of protected areas and IBAs in protection of farmland birds in Serbia

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Farmland bird populations are declining throughout Europe. The cause is considered to be rapid changes in agricultural practice, including intensification and land abandonment which result mostly from Common Agricultural Policy. These changes can occur in non EU member states as well, but at a lower level, since these are the countries that still apply traditional practices. To protect declining bird populations living in farmland, detailed knowledge on both species and communities is necessary. Population and distribution knowledge on the farmland species in Serbia is not enough, which can be an obstacle in perceiving the role of protected areas and defining the IBA. We used Maxent, a presence-only modelling method, to build species distribution models and to estimate effectiveness of network

of protected areas and IBAs in protection of six farmland bird species (*Motacilla flava*, *Sylvia nisoria*, *Sylvia communis*, *Lanius collurio*, *Emberiza hortulana* and *Miliaria calandra*) in Serbia. Gap analysis were conducted comparing percentage of suitable habitat inside and outside the protected areas and IBAs. Our results showed that current distribution of IBAs and protected areas in Serbia is not effective in farmland birds' protection because suitable habitats are mostly located outside the protected areas. Portion of habitats suitable for chosen species within protected areas varied between 1.41 and 2.72%. Within IBAs we found between 5.33 and 10.34% percent of total surface of chosen species suitable habitats. In conclusion, priority should be given to establishing a programme of agri-environmental measures in Serbia, which includes traditional practices, in order to conserve farmland biodiversity, as well as expanding borders of the IBAs and Protected Areas.

Keywords: farmland birds, suitable habitats, protected areas, IBA, gap analysis, Maxent

Landscape connectivity in river Bregalnica watershed - determining the relative importance of rural landscapes in terms of European wildcat conservation

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Landscapes are a mosaic of anthropogenic and natural ecosystems shaped as a result of long lasting interaction between humans and nature. At present, preservation of harmonious coexistence of man and wildlife within landscape(s) is of particular importance. In order to sustain the "harmony", landscapes should be managed to preserve their function to facilitate movement between resource patches. This is especially important in rural landscapes where the human impact on the landscape character is visible but still extensive, allowing it to serve as a link between "natural" landscapes that function as a core areas for wildlife. In this regard, the goal of this study is to assess the relative importance of rural landscapes as a wildlife habitat and corridor in terms of European wildcat (*Felis silvestris silvestris* Schreber, 1775) conservation.

Seven rural landscape types have been considered comprising 18 landscape units in the watershed of river Bregalnica in eastern part of the Republic of Macedonia. The results show that only 3 rural landscape types host adequate and sufficient habitat area to serve as core areas for the European wildcat populations: Hilly rural landscape, Osogovo mountain rural landscape and Mountain rural landscape, with Osogovo mountain rural landscape holding the most important role in regard of core area presence and core area importance for connectivity. From all rural landscapes, the hilly rural landscape has the highest value in terms of corridor presence and corridor importance. Furthermore, when all landscape groups in the river Bregalnica watershed are considered, the rural landscapes group holds the second highest value in terms of both core area and corridor presence and importance, after the forest landscapes

group that are considered as a prime core area for the wildcat.

The results of this study in respect of functional landscape connectivity in the region of Bregalnica watershed will contribute significantly to the wildlife conservation efforts and integrated management of ecological network in the region.

Keywords: Bregalnica watershed, connectivity, corridors, European wildcat, rural landscapes

Identification of priority areas for protection with the use of an Ecological Sensitivity Map in Bregalnica watershed (Republic of Macedonia)

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Considering growing threats to biodiversity and limited financial resources we identified and categorized conservation hot-spots and natural and semi-natural ecosystems important for management using Ecological Sensitivity Map (ESM). This way we were able to prioritize areas where conservations measures need to be undertaken.

The study area is the river Bregalnica watershed and the Eastern Planning Region, covering 4663km² or 18.1% of the territory of the Republic of Macedonia. Compared to the rest of Macedonia, where about 9% of the total area is under some form of protection, the study area's nature is disproportionally unprotected, with only 5 existing Protected Areas (PA) with a coverage of 0.68 km² (0.01% of the area).

We have used a set of 18 indicators, 6 of which are structural or abiotic, correlated with the risk of losing habitats identity/integrity, and 12 indicators are related to biodiversity and habitat connectivity, correlated with the risk of losing habitats ecological value. The produced ESM is a raster with 1km² pixel size, calculated as the average of all indicator values and classified in 5 categories using natural breaks (Jenks) algorithm.

According to the ESM, high sensitivity area covers 13.9% of the study area, 23.9% is medium-high, 26.3% is medium, 24.3% is medium-low, and 11.6% is low sensitivity area. The map confirmed the importance of 18 areas proposed for protection by the Spatial Plan of Macedonia and the Representative Protected Area Network, and identified 17 new areas as priority for protection. The coverage of all PA proposals is 979 km², 752km² of which is inside the study area and covers 16.1% of it. The proposed PAs mainly overlap with high and medium-high sensitive areas, covering 35.2% and 23.8% respectively. Also, the high sensitive areas outside PA proposal territories can be used for identification of corridors and areas important for mitigation of climate change and fragmentation effects.

Keywords: Conservation, Bregalnica watershed, sensitivity, protected areas

Structural properties of agricultural and rural landscapes in river Bregalnica watershed

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The human contribution in determining the landscape character is most visible in agricultural and rural landscapes. This long-lasting human-nature interaction had a distinctive role in nurturing secondary anthropogenic habitats which are significant for the preservation of biodiversity. Continuously, the change in local people practices reflects upon the landscape structure and its pattern. Assessing and managing landscape structural properties is crucial in order to secure the landscape functionality, especially in a region where there are ongoing conservational efforts. Following, the aim of this study is to assess the structural properties of agricultural and rural landscapes in river Bregalnica watershed.

For this purpose types and coverage of land cover classes in landscape units of both landscape groups have been assessed. Structural properties were assessed by calculating of patch number, mean patch size, average patch perimeter, average patch shape, averaged distance between the patches of the same land cover class and total edge of patch. The results show that fragmentation levels from high in agricultural landscapes to moderate in rural landscapes clearly separating both landscape groups in their capacity to sustain biodiversity.

Keywords: landscape structure, agricultural landscape, rural landscape, river Bregalnica watershed

Contribution to the study of floristic features of Prizren Castle in Kosovo and impacts of anthropogenic factors

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The Prizren Castle, located in southern part of Kosovo is one of the most visited sites in Kosovo due to the combination of historic, cultural and natural values. In the area of Prizren Castle and surrounding zone, in the territory of about 15 ha, a detailed investigation of floristic composition is made during years 2013-2014, following by anthropogenic factors impacts on it.

During the research work were collected and determined 153 plant species have been collected and determined, altogether categorized in 120 genera and 41 families.

Most represented families with species are: Asteraceae with 23 species, Fabaceae with 17 species, Brassicaceae with 14 species, Lamiaceae with 13 species, Scrophulariaceae 10 species, while other families have had less than 7 species. From species recorded, 36 species belongs to the medical plants group.

Research has proven that these plant species belongs to 14 floristic elements, and according to

their floristic spectrum, the largest number of 26 species belongs to Euro-Asian floristic element, 10 species to the Sub-mediterranean element, 8 species to the Mediterranean element etc. The Castle territory itself is rich also with ruderal plants. In the surroundings of the castle, in the eastern part is identified Balkan endemic species *Scabiosa fumarioides*.

The aim of the research was to identify the plant species in and around the Prizren Castle thanks to the impacts on them. Threatening factors of the Castle floristic diversity are mainly economic and cultural interests of the area. According to the city development plan, the renovation of the Castle infrastructure is foreseen, including escalators in the area, which will undoubtedly lead to the destruction of natural habitats and with significant threat of disappearance of many plant species.

Keywords: flora, castle, plant species, impact, cultural

Comparative morpho-anatomical analysis of species *Crocus reticulatus* Steven ex Adam (Iridaceae) from Serbia

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This research gives an overview of morpho-anatomical variability of *Crocus reticulatus* Steven ex Adam in Serbia. Study included four populations from Serbia, three from Pannonian Plain (Tomislavci village, near the city of Bačka Topola - 110 m altitude; hill Titelski breg – 128m altitude; and mountain Vršački breg - 250 m altitude) and one population from eastern Serbia (Srbovo village, near the city of Negotin - 89 m altitude). Thirty individuals per population were investigated. Analyzed morphological parameters showed that the only difference between lowland populations was at the level of the bulbs (bulbs width and height, and bulbs fibres width). Listed parameters have the highest values in individuals from population of the Vršački breg. There are no significant differences between populations on the basis of measured anatomical characters except number of ribs (Vršac Hill and Srbovo populations have four ribs, while the other populations have two ribs). Also, there is significantly higher value of particular measured parameters (xylem area, phloem area, sclerenchyma area, parenchyma area, the height and width of mesophyll and epidermal cells) in leaf cross sections of population from Srbovo village. Some authors have already indicated that *C. reticulatus* species in Serbia should be separated into more taxa – *Crocus variegates* Hoppe & Hornsch (with northern distribution in Serbia - Pannonian Plain) and *Crocus danubensis* Kernd. et al., near the Danube River (Negotin). We will be focused to clarify this issue in future investigations. Our research will be expanded on the Carpathian region of Romania and Hungary to investigate the ecological factors of the habitats and to get a better insight in phylogeography of *C. reticulatus*.

Keywords: *Crocus reticulatus*, morphology, anatomy, leaf, cross section

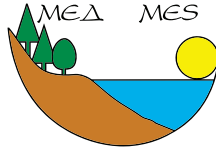
Distribution and valorization of batracho and herpetofauna in Kosovo and Metohia

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The amphibians and reptiles in the geographical regions of Kosovo and Metohia are widely spread and various. This is a result of many ecological factors that are influencing these two classes especially of the Mediterranean climate in the south western part and continental climate in the north eastern part of these regions. The data gathered in period of 10 years (from 2006 until 2015) can be considered as first conclusive study on these two classes that is covering these regions. The presents of 12 species of amphibians and 26 species of reptiles refers to the fact that the diversity of batracho and herpetofauna in these regions is rich and the results are referring on areas that can be considered as a biodiversity hot spots for these two classes. This results are first in obtaining the basic faunistic research for amphibians and reptiles that can be used for future actions in terms of protection and conservation.

Keywords: amphibians, reptiles, faunistic



Section 4

**HYDROBIOLOGY, WATER
MANAGEMENT AND
MONITORING**



Application of environmental oxygen and hydrogen isotopes in hydrology /Principles and Case study/

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Increased pressure on the environment and the natural resources all over the world threatens the economic potentials of the countries and hence their increased care especially for the available water resources. The resulting growing pressure in particular on groundwater resources requires detailed characterization and quantification of groundwater dynamics, from the time it recharges aquifers to its drainage into surface waters (rivers and accumulations).

The global hydrological cycle together with its driving force, solar radiation, forms the basic resource for primary biological production.

Most probably water has been in our solar system from the beginning and was formed by the thermonuclear fusion process that produced the elements of the periodic system and their compounds.

Isotope studies applied to a wide spectrum of hydrological problems related to both surface and groundwater resources as well as environmental studies in hydro-ecological systems are presently an established scientific discipline, often referred to as "Isotope Hydrology".

The nature of the isotopic applications is of course dictated by the specific character of isotopes, radioactive and non radioactive.

During the transition of compounds such as water from one phase to another, the concentration ratio of the isotopes of an element often changes, undergoes so-called *isotope fractionation*. Conversely, observing differences in especially the stable isotopic concentration ratios ($^2\text{H}/^1\text{H}$, $^{18}\text{O}/^{16}\text{O}$) informs us about certain geochemical or hydrological processes that took place.

Radioactive decay offers the possibility to determine an age, provided certain conditions are met. Noteworthy in this respect is the frequent application of dating groundwater –i.e. determining the time elapsed since the infiltration of the water- by comparing the ^{14}C or ^3H (tritium) activities in a groundwater sample with that of the recharge water. Moreover, also concentration differences of either radioactive or stable isotopes can also be used as a tracer.

Facing with the problem for determination the originity and mechanism of recharge of the water in Rashce Spring, observations of the tritium and stable isotope distribution within the local hydrological cycle touching the catchment area of the Rashce Spring. Based on the results obtained so far, a physical two components (deep ground water from the Polog Plane and Vardar river + local Zeden precipitation as a second component)mixing model for recharging of the Rashce Spring was created. By simulation the tritium output of the Rashce spring and comparing it with the measured value, MRT (Mean Residence Time) and the volume of the stored water within the aquifer located below the Zeden massif were calculated and respective values of 30 years and $4,872 \times 10^9 \text{ m}^3$, were obtained. These figures were supported by applying of Tritium/ Helium ($^3\text{H}/^3\text{He}$) dating method, as well.

Keywords: Principles of Isotope application in Hydrology, Tritium/ Helium ($^3\text{H}/^3\text{He}$) dating method, origin of water and mechanism of groundwater recharge/discharge, Rashce Spring/case study

Diatom response to the climate change during Marine Isotope Stage 11 in ancient Lake Ohrid

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Past interglacial periods are a focus for palaeoecological research due to their potential contribution to understanding ecological response to future climate change, including rapid climate change events. Marine Isotope Stage (MIS) 11 (ca. 425 – 365 ka) is of phase of low eccentricity and regarded as a relatively close analogue to the current, Holocene, interglacial. Marine core records extending back to MIS 11 are more common than terrestrial and lacustrine deposits. The latter can offer valuable insight into the impact of climate fluctuations in continental environments. Here, we analyse the palaeolimnological record of MIS 11 preserved in a long (ca. 2 Ma) lake sediment core from deep (ca. 288m maximum depth), ancient Lake Ohrid (Macedonia/Albania), the oldest extant lake in Europe. In the semi-Mediterranean climate, temperature change is likely to exert a stronger influence on ecology than shifts in available moisture. Other circum-Mediterranean lakes are far more shallow and dominated by lake-level response to humidity shifts so Ohrid is a key site for palaeoclimate research. Building on previous geochemical and palynological research, we focus here on ecological diatom response to limnological change driven by climatic forcing. Interestingly, during this warm period the diatom composition is dominated by medium to large-celled *Cyclotella* sp. nov, observed only in the fossil sediments, characterized by high morphological variability in the shape and in the valve ornamentation. This is opposite to more recent interglacials where small-sized *Cyclotella* species are more abundant. Planktonic species occurring in the modern lake are present at minor abundances, mainly comprising *C. fottii*, *C. ocellata* and *C. minuscula*. Based on the CONISS analysis, three diatom assemblage zones (ODAZ) were identified. In the ODAZ1 (425–411 ka), facultative planktonic and benthic species have higher abundance compared to the two following zones. Taxa with higher trophic preferences (*Stephanodiscus* spp.) are present. ODAZ2 (411–377 ka) is marked by an increasing trend of *C. minuscula* and *C. ocellata* at the expense of *C. sp. nov*, and punctuated by two distinct peaks of the small (3–7 µm) planktonic *S. minutulus*. Zone ODAZ3 (377–365 ka), which corresponds most probably with the stadial MIS11b, is characterized by very low abundance of *C. ocellata*, while *Cyclotella* sp. nov. has a maximum abundance of > 60 % at ca. 370 ka. In general the record exhibits marked shifts in diatom composition which probably is a result of temperature changes and nutrient availability. The first increase in T (ca. 417 ka) resulted with a change in the diatom community, where *C. ocellata* and *C. minuscula* appear, this is correlating with the highest TIC concentration, implying highest productivity. The latter species react rapidly to tephra deposition, here noticeable with maximum abundance ca. 395 ka. Further comparison with the pollen data and other geochemical proxies will provide more detail insight of the diatom response and ecology of the lake itself.

Keywords: diatoms, climate change, MIS11, lake Ohrid

Lake Cerknica – The ecosystem driven by water level fluctuations

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Lake Cerknica is intermittent lake located at karst polje Cerkniško polje. Water level fluctuations present a physical factor, which is of vital importance for the lake, influencing the abiotic and biotic processes. These promote or suppress the growth and development of vegetation, litter decomposition and even fungal colonisation of plant roots. The most important consequence of the intermittence is the exchanging of oxic and anoxic conditions in the lake sediments. Water level fluctuations create a variety of habitats with diverse communities. Habitats are delineated by duration and extent of flooding and soil properties. The variety is not only spatial but also temporal, resulting in a specific vegetation pattern. The life histories of organisms in the lake are intimately coupled to the periodicity of water regime. The success of plants in this outstanding environment depends mainly on their potential to overcome water level fluctuations i.e. morphological, physiological and biochemical plasticity, cosmopolitanism and as well as reproductive flexibility. Many species exhibit amphibious character. In Lake Cerknica the majority of shallower areas are colonised by the cosmopolite species *Phragmites australis* and some other wetland species. The water level fluctuations as a permanent disturbance are also a protection against alien plant species that have spread in Slovenia in the last few decades.

From the point of view of resilience of the ecosystem function and its role in the landscape it is crucial to preserve the natural water regime, to prevent pollution in the watershed of the lake and to increase the self-purification efficiency of the tributaries by remediation measures.

Keywords: intermittent lake, drainage, flooding, plants, decomposition

Studies on eucrenal-hypocrenal zonation of springs along the river mainstream: a case study of a karst canyon in Bosnia and Hercegovina

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The canyons in Dinaric karst are known to harbour a diverse aquatic fauna living in springs along the river mainstream. However, the knowledge on the ecology of these springs and also on the springs in mountainous areas of the Balkan Peninsula in general is poor. This study is focused on the macroinvertebrate assemblages of three different types of springs along the Cvrcka River (Republic of Srpska, Bosnia and Hercegovina). The aim of this study was to check whether existing criteria for regional spring zonations in Central Europe are applicable also for riparian springs with short springbrooks bordering high order streams. The macroinvertebrates were collected seasonally for one year at two different distances from the source: at the source, and approximately 2-3 m from the source („springbrook“). At the spring sources, we found 59 species and higher taxa while in the

springbrooks 61 species and higher taxa were recorded. Diptera represented the most abundant taxon, followed by Amphipoda and Trichoptera. No strong trend for the Margalef's index for the spring source and springbrook was detected, while the Shannon's diversity index increased in the springbrook of the studied springs. Our analysis did not prove significant differences between the macroinvertebrate assemblages from the source and springbrook. Based on faunistic data we suggest that small riparian springs with a short outflow likely do not exhibit true spring zonation but may show a "quasi-zonation" defined as a possible hidden differentiation between source and spring outflow. Our study shows that criteria for spring zonation are not suitable for riparian springs bordering high order streams.

Keywords: macroinvertebrate, springs, Cvrcka River, Republic of Srpska, Bosnia and Herzegovina

Factors affecting distribution pattern of chironomidae larvae (Diptera: Chironomidae) in Bregalnica River, Republic of Macedonia

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The degradation of the water quality has become a major environmental concern due to continued anthropogenic pressure. To protect rivers, various methodologies have been developed to assess their ecological status. Several studies, have demonstrated that Chironomids (non-biting midges) are responsive to environmental changes with different species possessing different response. We investigated the relation between larval chironomid assemblages and heavy metal and organic pollution in Bregalnica River. Samples of Chironomidae larvae fauna were collected during 2007-2008 at two sampling sites of the river, downstream of the Sasa lead-zinc mine and the city of Štip. Chemical and physical parameters of water and sediment were also examined. Results showed significantly high density of Ortocladinae species (which prefer Cd and Pb rich sediment) at the first site, as well as Chironomini species (tolerant to organic pollution) at the second site. Statistical analyses confirmed that sediment characteristics have a more significant impact on Chironomidae larvae than the analyzed water parameters. The findings of this study indicate that non-biting midge larvae displayed a strong pollution response to both organic and heavy metal contaminations, making this group an effective and reliable indicator for the ecological status assessment of the water bodies in R. Macedonia.

Keywords: Chironomidae larvae, Bregalnica River, environmental variables, indicators, pollution

Long-term monitoring strategy of water through the target organs of fishes from marine and freshwaters

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Based on the global changes of the environmental pollution, different countries are trying to make a strategy for monitoring those disorders that appear in different ecosystems. An ecosystem approach to fish and wildlife conservation means protecting or restoring the function, structure, and species composition of an ecosystem while providing for its sustainable socio-economic use.

As experimental animals for the realization of this study were used of the *Cyprinus carpio* like a control fish and the experimental fish of marine and freshwaters caught in several locations, through “electroshock” method, using the CYBER POWER generator. The EROD enzyme activity was measured with spectrofluorometers (SHIMADZU RF-1501), according to the method applied by Burke and Mayer (1974), for excitation/emission wavelengths setting 510nm/585nm. The staining was done according to the method of H&E and protocol Harris, while the microscopic examination of lesions is done through the fluorescence optical microscopy, with digital cameras and software (NIKON Eclipse 80i).

The results obtained from the analysis of the activity of the EROD enzyme, in the control and experimental fishes indicate an increasing activity of EROD in the presence of pollutants and xenobiotics. As well as the results from histopathological changes of the liver parenchyma and gonad sections of the experimental fish include vacuolization of hepatocytes; hypertrophy of epithelial cell nuclei of biliary duct; macrophagous aggregates; degeneration hepatocytes; sinusoidal expansion.

Contemporary or modern trends based on the development of the technology in the last years are representing a threat for the earth planet in general and for the water environment as a specific part. The strategy of global monitoring on flowing waters, like the river, the lake, and the sea water, can be forwarded on a permanent way through biological markers (bio enzyme markers) which make that pathological difference in the liver of fish in the water environment.

Keywords: biomarkers, environmental pollution, histopathological changes.

Macrophytes as water quality elements - Implementation of WFD in Slovenia

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Macrophytes are one of the biological elements required by the EU Water Framework Directive (WFD) (Council of the European Communities, 2000) for assessing the ecological status of rivers and lakes. Species composition and abundance reflect the quality of ecosystems. Slovenia cooperated in three Geographical Intercalibration Groups with macrophytes: Mediterranean (Med)-GIG, Eastern Continental (EC)-GIG and Alpine (Alp)-GIG. In the (Med)-GIG and (EC)-GIG Slovenia cooperated with rivers and in the (Alp)-GIG with lakes. For assessment of ecological status of rivers we have developed an index RMI – River Macrophyte Index. We combine RMI index with phytobenthos indices to classify river stretches into certain ecological status. In Med-GIG and EC-GIG intercalibration with other member states was successful. In addition, in the frame of the (Alp)-GIG we calibrated SMILE, Slovenian macrophyte-based index for lake ecosystems. Macrophyte data were obtained using the transect method. Index SMILE is also combined with phytobenthos index to classify lakes into certain ecological status. Huge amount of data that were used for the developments of both indices were gained from national monitoring programme, different projects as well as from diploma, master and doctoral works.

Keywords: ecological status, macrophyte composition and abundance, lakes, rivers

Epiphytic macroinvertebrate assemblages in Trskovača pond (Platičevo, Serbia) under restoration management

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Aquatic invertebrates have been used as indicators of habitat restoration owing to their ecological preferences, sedentary nature and long life cycles. Epiphytic macroinvertebrate assemblages comprise freshwater populations associated with macrophyte stands. Trskovača pond, situated in Srem, southern part of Pannonian plain, is the remnant of once larger wetland area which has been highly modified in the past by intensive agricultural and melioration activities leading to complete drainage of the pond itself. After spring floods in 2014 the pond ecosystem was re-established, and maintained owing to active control of the water level in the adjacent irrigation canals. In order to evaluate the effects of habitat restoration as well as to assess current ecological state of Trskovača pond, we started monitoring epiphytic macroinvertebrate assemblages. The research was conveyed during medium high hydroperiod in summer 2015 and 2016. Samples were collected using standard sampling net for qualitative analyses. The results showed unusually high diversity both in the first and the second year of sampling. The taxa

richness of epiphytic macroinvertebrate assemblages comprised up to 20 families. The most abundant were species with high level of mobility and dispersal capacity belonging to families Corixidae, Pleidae and Notonectidae (order Heteroptera). Presence of non-insect taxa: snails (mostly presented as *Radix labiata*) and crustaceans (*Asellus aquaticus*), indicated stable aquatic conditions and the potential for establishment of diverse freshwater communities leading to restoration of this wetland area.

Keywords: freshwater monitoring, aquatic invertebrate fauna, restoration success, lowland wetlands

Seasonal variation of metal concentrations in different tissues of the European chub (*Squalius cephalus* L.)

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Metals present a serious threat to aquatic organisms because of their toxicity, persistence and bioaccumulation. They are continuously released into the aquatic environment from the geological and anthropogenic sources. The aim of this study was to analyze the metal and metalloid levels (using ICP-OES) in liver, gills, gonads and muscles of European chub (*Squalius cephalus* L.). The European chub is a ubiquitous fish species displaying a wide niche, feeding on a low trophic level and widely distributed all over Europe. The total of 65 specimens were from the rivers Pestan and Beljanica, which belong to the Kolubara basin that is subjected to intensive exploitation and refining of lignite. The sampling at Pestan (8 months), and Beljanica (10 months) was conducted from October 2011 to September 2012.

Significant differences in the concentrations of elements in different seasons were found at Pestan in: gonads (As, Ba, Zn, Hg), liver (Cr, Mn, Zn, Hg and Sr), gills (Mo, Zn, Hg), muscle (Zn, Hg), while at Beljanica in: gonads (Mn and Sr), liver (Cu, Hg), muscle (Cr, Hg), gills (Ba, Cu, Mo, Sr, Zn). The accumulation of elements was highest in the gills and lowest in the muscles. Both rivers showed a similar pattern of accumulation of elements and seasonal changes. Increased accumulation of elements was observed during the summer. Zn and Hg showed a pronounced seasonal variation in all tissues. Principal component analysis (PCA) singled out Hg as the element with the highest concentration in muscle during autumn. Canonical discriminant analysis (CDA) is, in relation to the total content of the elements in the tissues, demonstrated total separation of the gills and partial separation of the liver. The accumulation of elements was highest in the gills and lowest in the muscle.

Keywords: ICP-OES, fish tissues, metals and metalloids

Environmental predictors of the pondweeds occurrence in the Middle River Danube in Serbia

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Pondweeds play significant role in the functional diversity and integrity of freshwater ecosystems. The main objective of this study is to understand the relation between pondweed species and their environment in large, alluvial river such as Danube in its middle course.

The survey of pondweed species assemblages and environmental parameters was done in 1082 contiguous survey units (SUs), between river km 1433 and 846 of the Danube River main channel and in the backwaters with permanent surface inflow. Environmental parameters were grouped into: hydrological, morphological, biological and the type of prevailing land-use along the bank of the SU. Data was analyzed using PCA, RDA and pRDA.

We have recorded: *Potamogeton x angustifolius*, *P. crispus*, *P. gramineus*, *P. lucens*, *P. natans*, *P. nodosus*, *P. perfoliatus*, *P. pussilus*, *Stuckenia pectinata*. 25.9% of species variability is explained by analyzed environmental variables. The effect of each group of environmental variables is highly significant, the most important is hydrological, explaining 18.3% of variability in species data. The distance from the dam has strongest marginal effect. Forward selection included seven variables into the RDA model which explains 25.7% variability in species data. Species are divided in two groups: a) pondweeds occurring in reservoirs Đerdap I and II, growing on fine sediment, in SUs with coarse bank material (*P. natans*, *P. nodosus*, *P. perfoliatus*, *P. pussilus*); b) pondweeds growing in the main river channel, tolerating higher flow velocities, in SUs with abundant submerge vegetation and with arable land along the banks (*P. angustifolius*, *P. crispus*, *P. gramineus*, *P. lucens*, *S. pectinata*).

These results highlighted some interesting problems for the future studies, such as to explore competitive relations between pondweeds and other hydrophytes, and the influence of arable land on aquatic vegetation.

Keywords: Potamogeton, Stuckenia, multivariate analyses, variation partitioning

Assessment of macrozoobenthos and environmental quality in the Albanian part of Macro Prespa Lake

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The aim of this study was the assessment of species composition and quantitative characteristics of the macrozoobenthic community in the Albanian part of Macro Prespa Lake, and of the environmental state of the lake, based on benthic macroinvertebrates as indicators.

Sampling has been carried out in two sites, Gollomboç and Liqenas, in October 2013, from 0,5

m to 10 m depth, using the multihabitat transect method. A “kick and swipe” net has been used for sampling in 0,5 m depth, while in 2 m, 4 m, 6 m and 10 m the samples were taken by a box-corer.

A low species richness and low abundance was recorded in the sampling sites. Gastropods were the predominant group regarding the number of species and abundance.

Macrovegetation, especially the algal cover of *Chara*, plays a very important role in the species composition and quantitative characteristics of the macrozoobenthic community in the studied area. The macrozoobenthic community had a low stability and its structure was assessed to be in a “moderate” to “poor” state, based on some stability and diversity indexes.

The environmental quality of the lake was predominated by the “poor” to “bad” status, after the WFD categorization. Environmental indicators based on benthic macroinvertebrates show a tendency for lake Eutrophication, nutrients’ enrichment and increased organic pollution.

However, the lake is still a shelter for many benthic macroinvertebrate species of international concern and of interest for conservation, including endemic species and globally threatened species.

Keywords: benthic macroinvertebrates, environmental quality, Prespa Lake.

Alien species of Skadar Lake in Montenegro

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In this paper the inventory of non-native vascular flora of Skadar Lake is presented.

Skadar Lake is located on the Montenegrin-Albanian border. Montenegrin part: National Park since 1983 (IUCN Management Category II); since 1995 Lake was recorded in the Ramsar List.

The inventory has been created on the basis of unpublished data from field observations, data from literature as well as data from herbaria.

The inventory lists 145 non-native species and subspecies which presents about 10 % of the total flora of Skadar Lake and about 4 % of the total flora of Montenegro.

The non-native flora of Skadar Lake is listed with information on the taxonomic position (species and subspecies), life-form, origin, residence time status (archaeophytes vs. neophytes) and invasive status (casual, naturalized, invasive).

According to its current invasive status the number of 59 (40,7 %) naturalized non-invasive taxa, 51 (35,2 %) invasive and 35 casual (24,1 %) taxa were registered in the alien flora of Skadar Lake. The alien flora of Skadar Lake consists of 49 (33,8 %) archaeophytes and 96 (66,2 %) neophytes.

Keywords: Alien species, invasive species, Skadar Lake, Montenegro

Drip water in the cave Slatinski Izvor – hydrochemical properties and influence on the Slatinski Izvor spring during low waters

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The Slatinski Izvor cave is located in the Poreče basin of West - Central Macedonia. The Slatinski Izvor spring serves as one of the cave’s entrances of this spring cave, and represents the main outflow of groundwater from the karst aquifer. The recharge of the karst spring is a combination of both, allogenic and autogenic recharges.

Drip water in the cave Slatinski Izvor was analyzed in the period of low water between December 2011 and October 2013. Water was sampled from three stalactites. The measurements of the temperature and conductivity, and the results of hydrochemical analysis indicate that all water samples have very similar physical and chemical properties. Four methods (Piper diagram, Stiff diagram, Chadha diagram, and D’Amore diagram) were applied in order to determine the hydrochemical properties of the water. The major cations in the water were Mg^{2+} , Ca^{2+} and Na^+ , and anions were HCO_3^- , SO_4^{2-} and Cl^- .

Drip water from the cave contributes to the recharge of the spring Slatinski Izvor. During low waters spring water tends toward larger contribution of Mg^{2+} which is characteristic for drip water and indicates larger share of autogenic recharge.

Keywords: karst drip water, cave Slatinski Izvor, hydrochemical analysis, Republic of Macedonia.

Application of CE-QUAL-W2 model to nutrients dynamics simulation in Lake Prespa

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The lake Prespa is modelled to simulate the nutrients dynamics using the CE-QUAL-W2 model, which is a carbon-based, laterally averaged, two-dimensional water-quality model. The modelling effort was supported with data collected in the field for a 2-year period. At monitoring site, nutrients profile measurements were taken from the surface through the water column. The validation process consisted of comparisons of predicted in-lake concentrations to those observed during the monitoring period, which extended from May through November 2013 and March through October 2014 with a 2-week offset. The focus for evaluating the model validation was given to three constituents of nitrogen and two constituents of phosphorus: nitrate plus nitrite, ammonia, total nitrogen, orthophosphorus, and total phosphorus. The sample data for each nutrient parameter were statistically analyzed on the basis of the four metrics of mean errors (ME), mean absolute errors (MAE), root mean square errors (RMSE),

and relative error of the mean in percentage (REM), using measured inputs of nutrients. The taken results showed that the CE-QUAL-W2 model was able to accurately simulate nutrients, with relative error of the mean less than 10%. Model results revealed that model simulated concentrations of key nutrients indexes matched well with the measured values. The results and conclusions from this study are not intended for use on Prespa Lake alone. The concepts of model development can potentially be applied and also broaden its usefulness to other systems.

Keywords: CE-QUAL-W2 model, nutrients dynamics, Ohrid Lake, simulation

The use of the Index of Trophic Completeness – ITC as an indication of water quality along the river Nišava river (eastern Serbia)

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Environmental anthropogenic disturbances result in changes of trophic structure in macroinvertebrate communities. Those changes can be detected using the ITC - index of trophic completeness. We chose this particular index to overcome the problems of using conventional indices in biological assessments of rivers, such as limitation to distinct geographical regions. Our goals was to determine quality of water along the river course using ITC and to determine minimal distance of the self-purification processes to become effective. Macroinvertebrate samples and physicochemical data were analyzed for 12 localities along the Nišava river in Southeastern Serbia during a one-year period. Even-numbered localities are positioned downstream and odd-numbered upstream of wastewater discharge points. The ITC – index was the lowest at the localities 10 (16,5) and 4 (17,7). Locality 10 is site with the lowest average concentration of oxygen (6,5 mg/l) and locality 4 is site with the highest total nitrogen concentration (0.34 mg/l) and the highest values of BOD₅ (4,16 mg/l). According to ITC along the river we detected 3 classes of water quality: high, good and moderate. In the high class of quality belongs only locality 11. In the good quality class there were three subclasses (subclass one-localities 2, 3, 5, 7; subclass two-locality 8; subclass three-localities 6, 9). In the moderate class of quality there were two subclasses (subclass one-locality 12; subclass two-localities 1, 4, 10). We concluded that the minimal distance of the river self-purification processes to become effective is at least 8 km along investigated river.

Keywords: macroinvertebrate communities; index of trophic completeness; water quality

Accumulated amounts of water in Republic of Macedonia

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Republic of Macedonia is located in temperate zone, generally exposed to south southeast, with higher temperature influences thus receiving less precipitation. That expresses the necessity of accumulated amounts of water.

The main aim of the paper is analysis of accumulated amounts of water. By implementing an inventory of all natural and artificial accumulated amounts of water, the work is focused on their distribution and use, also evaluation of water necessity. The contribution of the paper refers to identification of the current state of waters, emphasizing on accumulated waters, valuation of water necessity and the impact on geographical and ecological environment.

Keywords: Republic of Macedonia, natural lakes, artificial lakes, accumulated water, geographic environment, environmental environment.

Assessment of the trophic state of Strezevo reservoir

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“Strezevo” reservoir is one of the largest reservoirs in Macedonia which has multipurpose utilization. The reservoir provides water for irrigation, water supply, water utilities and other needs, the production of electricity, supplying technological water for industry etc.

Subject of this work is to determine the trophic state of accumulation Streževo based on physico-chemical and biological parameters.

During August 2009 to August 2011 collection of water samples was made from seven measuring places with seasonal dynamics: the flow of river Semnica into the reservoir, and vertical profiles of central part and part of tower intake (0,5 m, 8 m and 20 m depth).

In order to assess the trophic state of reservoir, it was investigated the following physical-chemical and microbiological parameters, transparency of water, organic matter, total phosphorus, coliform and heterotrophic bacteria, chlorophyll a concentration. The results received from research indicated that the water quality has got changeable character which depends on seasonal changes and anthropogenic activity.

According to the concentration of total phosphorus and trophic state index, the water has oligotrophic-mesotrophic character. According to the heterotrophic bacteria accumulation “Streževo” in I and II class, based on the coliform bacteria indicate the sanitary aspect, the accumulation is mostly class II or II - III class in separate parts of summer 2009. The concentration of chlorophyll a has seasonal dynamic, with maximum values in summer. Due to the intermediate value, the concentration of

chlorophyll a, the water of Strezevo reservoir has mesotrophic character.

Using Carlson trophic state index for determining trophic state of lakes, reservoir Strezevo belongs to the group of oligotrophic - mesotrophic lakes. According to the OECD classification and parameters: total phosphorus, chlorophyll a and transparency, water from the reservoir Strezevo in the period 2009/11, has mesotrophic character.

The obtained results of the research indicate the necessity of continued and complex research because of the control, realising the situation timely and take an appropriate measures to protect the water quality of this multipurpose aquatic ecosystem.

Keywords: “Strezevo” reservoir, trophic state, physico-chemical parameters, heterotrophic bacteria, coliform bacteria, chlorophyll a, Carlson trophic state index

Preliminary investigation of the ecological state of Lištica and Radobolja rivers, Bosnia and Herzegovina

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Lištica and Radobolja are karst rivers, known for its beauty, as well as habitats of specific aquatic flora and fauna. Since ancient times the waters of these rivers have been used for water supply, agricultural irrigation and recreation, and more recently their ecosystems are increasingly exposed to various types of anthropogenic pressures. During 2016 we started investigations of the Lištica and Radobolja rivers in order to assess the ecological status and the quality of their waters. Studies included three biological quality elements: macroinvertebrates, phytobenthos and macrophytes, as well as physical and chemical parameters. For biological and physico-chemical elements we applied a methodology based on European standards of water quality (Water Framework Directive, 2000/60 / EC) in accordance with national protocols sampling (the Water Law of F BiH “Official Gazette of BiH”, No. 70/06). Biological samplings were carried out according to the ISO 5667 regulations and AQEM methodology, at four sites along the longitudinal profiles of both studied rivers. This paper presents the preliminary results of the research, which are the basis for planning further activities for sustainable use of these areas.

Keywords: water quality, macroinvertebrates, phytobenthos, macrophytes, physico-chemical parameters

Distribution of macrozoobenthos in Rijeka Oskova, tributary of river Spreča (Bosnia and Herzegovina)

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Yearlong research of macroinvertebrates from river Oskova which is a left tributary of river Spreča showed a relatively big variety of species. Presence of 83 taxa with a total of 153 individuals was determined during the research period from April 2014 to May 2015. Sampling was done by the method “kick sampling”. The study was conducted on six sites of which the first two were Velika Zlaća streams and Krabanja stream who merge and create river Oskova up to the mouth of this river where it merges with river Spreča. The largest number of individuals of macroinvertebrates was found on the third site (Zlaća), while the smallest number of individuals was found on the fifth site (area around settlements Višća). Studies have shown that the number of species decreases from the source area, where 51 species are recorded, to the mouth of Oskova where 30 species of macroinvertebrates were recorded. The smallest standard deviation (6.007) have macroinvertebrate community from stream Krabanja and the biggest (13,658) have communities that live on the bottom of the mouth of river Oskova. The standard error and the standard deviation have the lowest value on the second site and the highest at the sixth site. The diversity of macroinvertebrate communities in the districts of research has been shown by the Shannon - Weaver’s index of diversity. The index of diversity from the site Velika Zlaća (L1) and Krabanja (L2) show the maximum value (1.45) which means that the macroinvertebrate communities from these two sites were the richest. Lower values of diversity were registered on the site mouth of Oskova (L6), which amounts to 1.05. All obtained index values were between one and two which indicates a high pollution of water. The results of cluster analysis show a degree of similarity of macroinvertebrate communities from the locality Velika Zlaća and stream Krabanja. Smaller index of similarity show communities caught at the site 4 (settlement Ježevac) and the mouth of Oskova. A smaller similarity index have the samples from the third site (Zlaća) and the lowest index have the isolated macroinvertebrate communities from the site Višća. Testing of water quality was performed using the biotic and saprobic index. Based on the value, we can differentiate headwaters of river Oskova which belong to the category of clean waters and the lower course with a steep decline in quality with the presence of organic origin pollutants.

Keywords: Macrozoobentos, saprobity, distribution, water quality.

Seasonal zooplankton dynamics of Dojran Lake (Republic of Macedonia)

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The taxonomic composition of zooplankton community in Dojran Lake were studied during the spring - autumn season 2015 in order to understand the differences in the abundance of different taxonomic groups. In total, 29 taxa were identified, of which 19 representatives from Rotifera, 4 from Cladocera and 3 from Copepoda. Bivalvia and Chaoboridae larvae were presented with only one representative each. From quantitative point of view, Cladocera (47,7%), Copepoda (31,2%) and Bivalvia (19,7%) were the most numerous in the zooplankton community. In compare with previous research on Dojran Lake contribution of Rotifera and Chaoboridae larvae significantly decreased (1%). Although the composition of the zooplankton changed during the seasons, results showed that water flea *Daphnia cuculata* and calanoid copepod *Eudiaptomus gracilis* were present the whole period. The maximal density of the *Dreissena presbensis* larvae was evident in the summer period, while it completely disappeared from the zooplankton community during the spring and autumn period, which corresponds with the life cycle of these mollusks. The most significant changes in the taxonomic composition occurred during autumn period starting with the presence of the cladoceran *Diaphanosoma brachyurum* reaching higher density, the detection of the larvae *Chaoborus crystallinus* in the open water and the maximal presence of transparent predatory water flea *Leptodora kindtii*. In terms of rotifers, Lake Dojran is characterized by a predominance of periphytic or littoral elements and fewer plankton species. This feature can be assigned to the lack of definite pelagic habitats, shallow nature and growth of aquatic macrophytes. The simultaneous presence of several species of the genus *Brachionus* and their high abundance is a good indication on higher trophic status of Lake Dojran.

Keywords: zooplankton, Dojran Lake, qualitative composition, quantitative composition, eutrophication.

Aquatic macrophytes have significant role in shaping phytoplankton community

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Shallow aquatic ecosystems represent suitable entities where the relationship between aquatic macrophytes and phytoplankton community can be monitored. Over a wide range of intermediate nutrient levels, two alternative states can exist in these ecosystems: one with clear water and macrophytes dominance, and a more turbid one with algal dominance. Each of these equilibria states is stabilized by

buffer mechanisms, but the central spot is the interaction between submerged vegetation and turbidity.

The object of our study was the Zasavica River, shallow aquatic system, which is a part of the Special Nature Reserve “Zasavica” (Serbia). It can be characterized as both standing and slow-flowing water according to its prolonged retention time in some periods. On the other hand, dense and diverse macrophytes populations develop from April to October in this river. Considering the scarcity of the data related to phytoplankton in the Zasavica River, the aim of this study was to investigate its structure and dynamic.

Phytoplankton samples were taken monthly from December 2012 to November 2013 at two localities. Standard literature was used for qualitative analysis, while quantitative analyses were done by using Utermöhl method. Biomass of phytoplankton taxa was determined by geometrical approximations.

Total of 392 phytoplankton taxa from 8 divisions were recorded, but only 69 of them had biomass $\geq 2\%$ at least in one sample. The highest recorded abundance and biomass were 15524 cells/ml and 3716 $\mu\text{g/l}$, respectively. Although diatoms were dominant phytoplankton component in most samples according to biomass, flagellated taxa made the second important group with significant number of mixotrophs, which is expected in macrophytes dominated systems. The macrophytes dominance in the Zasavica River keeps the water clear by maintaining small phytoplankton abundance and biomass with extraordinary richness of taxa and also has significant role in shaping phytoplankton structure by promoting flagellated mixotrophs.

Keywords: the Zasavica River, shallow aquatic ecosystems, richness

Detection of SVCV in tissue of common carp, *Cyprinus carpio* L. through ELISA techniques

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Spring viraemia of carp is a viral disease that affect mainly *Cyprinidae* family and is listed as a notifiable disease by the World Organisation for Animal Health (OIE). So the diagnostic procedures of the virus, *Rhabdoviruscarpio*, that caused spring viraemia of carp disease should be based on the OIE guidelines (Office International des Epizooties 2009). SVCV is serologically distinct from the other fish rhabdoviruses. Therefore one of the technique used for the detection of SVCV is ELISA. The samples, *Cyprinuscarpio*, were collected from different region of Albania, from Lake Shkodra, Prespa Lake, from aquaculture pond in which common carp is cultivated etc. Internal organs, liver, spleen, kidney and encephalon tissue were used for the detection of SVCV. After the homogenization of the organs they were centrifuged at 3000 xg for 15 minute at 4°C. We made a serial of dilution 1:10 of each supernatant which are used for the infection of EPC cell line, L-15 medium with 2% fetal bovine serum. Then subcultivated in order to isolate the virus. Then the plate is frozen and thawed for three cycles in order to release the virus from the EPC cell infected. This plate is tested with ELISA sandwich assay. For the test we used 96-well microtitration plates, BIO X k 275/1, sensitized by specific antibodies for SVC virus. A positive control antigen is provided with the kit so as to validate the test results. In every well of the plate, except for the positive control one, we add 100- μl aliquots of the supernatant taken

from the EPC cell plate infected. The signals recorded from spectrophotometer show a negative result for the 69 samples of common carp.

Keywords: SVCV, ELISA sandwich, L-15 medium, FBS, *Cyprinus carpio*.

The establishment of the invasive blue crab *Callinectes sapidus* Rathbun, 1896 in the Lagoon of Vain (South-east Adriatic Sea, Albania)

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The blue crab *Callinectes sapidus* Rathbun, 1896 is considered as an invasive species in the Adriatic Sea and has been largely distributed along its coast during the recent years. Based on personal communications with the local fishermen of Vaini Lagoon, the blue crab appeared in the Vaini area in 2010. The presence of the blue crab *Callinectes sapidus* has been largely recorded in the Lagoon of Vain, especially in its northern part, at the mouth of Drini river. The aim of the present study is to provide data on the distribution, assessment of the abundance, the structure and biometric characteristics of the blue crab population in Vaini Lagoon.

Samples have been collected in March and October during 2014-2015. Besides direct observation and samples' collection in the study area, questionnaires have also been distributed to the local fishermen with the purpose of gathering information about the presence of the blue crab, assessment of its state and its possible impact on the other populations in the Vaini Lagoon. Based on biometric measurements most of these crabs can be considered as mature. Taking into account the high abundance and frequency in the fishing nets, as well as our records of the presence of juveniles and ovigerous females, the population of the blue crab *Callinectes sapidus* can be considered as established in Vaini Lagoon.

Keywords: blue crab, invasive species, Vaini Lagoon, biometric characteristics.

Comparative review of floristic structure in phytoplankton, phytobenthos and periphyton communities in shallow urban lake (Sava Lake, Serbia)

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Sava Lake is a shallow urban reservoir used for multiple purposes, exposed to intense anthropogenic pressure. Considering that cyanobacteria and algae represent a significant component of phytoplankton, phytobenthos and periphyton, that are all good bioindicators, it is interesting to

compare their floristic structure. The aim of this study was to evaluate diversity, as well as similarities between phytoplankton, phytobenthos and periphyton communities. All samples were collected simultaneously, weekly, from middle of July until the middle of September 2014. Phytobenthos was collected by scrubbing from cobbles (littoral), periphyton was sampled from artificial substrates (glass, ceramic, willow and yew tiles) previously deployed in the pelagic zone, and for phytoplankton samples planktonic net and Ruttner bottle were used. Identification of algal and cyanobacterial taxa in samples was done using standard literature, Carl Zeiss AxioImager M1 microscope and digital camera AxioCam MRc5 with AxioVision 4.8 software. Total of 63 taxa of algae and cyanobacteria were recorded in phytobenthos, 87 in periphyton, and 177 in phytoplankton. According to the CCA, when the type of community was used as explanatory variable, the three groups of all recorded cyanobacterial and algal taxa were clearly distinguished. A high number of unique taxa were found in each group, and those found in two or even three groups represent a smaller portion. However, the formation of these groups was not as much influenced by physico-chemical water parameters, as it was the consequence of the preference of algal taxa to the specific type of community. Cluster analysis based on species composition confirmed a greater similarity between phytobenthos and periphyton in comparison to phytoplankton. Generally, all three investigated communities in Sava Lake are unique compared to each other and have great diversity potential, which changes slightly each week. Moreover, considering the pressure on this waterbody, monitoring of the tree communities should continue.

Keywords: Algae, cyanobacteria, bioindicators, monitoring, anthropogenic pressure

Temporal variability of diversity and biomass of tintinnids (Ciliophora) in the Boka Kotorska Bay (South Adriatic Sea)

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Microzooplankton, a group of heterotrophic and mixotrophic planktonic organisms between 20 and 200 μm in size, is one of the most sensitive indicators of changes in the marine ecosystem. Ciliates of the microzooplankton include tintinnids, a minority component ideal for studies of species distributions and diversity.

Although the microzooplankton composition and dynamic is known in most parts of the Adriatic Sea, some areas of coastal ecosystems are poorly researched. One of them is the Boka Kotorska Bay, a complex geomorphological structure that is influenced by a large intake of fresh water through numerous submarine springs and rivulets and partly open waters of the southern Adriatic Sea.

The objective of this study was to determine the taxonomic and community data of tintinnids. Their biomass, temporal and spatial diversity were analyzed monthly during 2013 at seven stations: six located at Boka Kotorska Bay and one at the open Adriatic Sea. The distribution of tintinnid assemblages was also related to major physical and biological variables. Results suggest high species diversity, especially at inner part of Bay with occurrence of new species. Likewise,

seasonal environmental factors such as water salinity, temperature and phytoplankton concentration exert an influence on the species composition and biomass of tintinnids.

Keywords: Ciliophora, Boka Kotorska, Adriatic Sea

Microbiological assessment of the Lakes Bistarac and Vidara water quality

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In the spring and summer of 2014, the change of the number of coliform and heterotrophic bacteria in specific localities of Lake Bistarac and Lake Vidara were observed and analyzed. Lake Bistarac was formed after the cessation of surface mining in the open pit, while Lake Vidara is the artificial lake in the area of Gradacac made in order to protect the industrial zone of flooding. Both of these lakes, placed in Tuzla Canton, are very attractive for tourist and frequently are used for swimming, fishing and recreation. Results of this study showed an increased number of coliform bacteria in both periods, and the presence of *Escherichia coli* as a direct indicator of fecal pollution, while, as an additional indicator of fecal pollution, *Citrobacter* and *Enterobacter* were identified. From the total number of analyzed water samples of the Lake Bistarac, 60% of samples belonged into to the second class, and 40% into the first class of water quality. Lake Vidara showed poor quality as the 80% of the analysed samples belonged to the second class and only 20% into the first class of water quality. For the microbiological assessment of the water quality standard microbiological procedures and methods were used. One of the main reasons for poor hygienic state of these two lakes is unregulated sewage network of the villages, restaurants and camping areas placed near the lakes. The increased number of fecal bacteria was found in the water of both lakes, which can be highly dangerous for the human population. Since both lakes are used for recreational purposes it is necessary to establish an appropriate monitoring system on the basis of which is possible to take measures and to protect and improve water quality.

Keywords: water quality, coliform bacteria, fecal pollution, hygienic assessment

Benthic cyanobacteria in streams of Mt Tara (Serbia)

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Understanding of the composition of the phytoenthos community can give useful indications about the status of the waterbody, as well as appropriate management strategies, thus WFD entails monitoring of the phytoenthos as an element of quality used for ecological status assessment. Generally,

in Serbia examination of phytobenthos includes mainly monitoring of diatoms through calculation of diatom indices. As a consequence, data concerning geographical distribution and biodiversity of benthic non-diatoms and cyanobacteria is deficient in last two decades. Although nowadays there is a growing awareness concerning health risks associated with toxic cyanobacteria, most of the attention is dedicated to planktonic taxa. However, benthic counterparts are also increasingly recognized as problematic, bearing in mind that they can produce a range of cyanotoxins.

Phytobenthos samples were taken from three streams (Batarski and Karklijski Rzav, Vrelo Perućac) in the spring and autumn 2007, 2012 and 2015. Also, in 2015 the additional samples were taken in summer and winter from Vrelo Perućac.

There has been no published data concerning these localities so far. In this study, species composition of the benthic cyanobacterial assemblages were investigated in order to expand the knowledge about their distribution in Serbian flowing waters, as well as to highlight the potential risk concerning the occurrence of potentially toxic species. Fifteen genera with 27 taxa were present in the three streams. Among identified taxa representatives of the order Oscillatoriales were the most common, while the orders Chroococcales and Nostocales were represented with only few taxa. Some of these taxa were detected as dominant in the phytobenthos community. The highest diversity has been observed in Vrelo Perućac. Some of this taxa (*Pseudophormidium radiosum*, *Cyanosarcina chroococcoides*) were recorded for the first time in Serbia, while *Tolypothrix distorta* was previously recorded only in epifitic community on Mt. Stara Planina nearly two decades ago.

Keywords: phytobenthos, diversity, geographical distribution

Microbial diversity of sediments from cave Vrelo

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Caves, with relatively limited nutrient in organic matter, stable and low temperatures, high humidity and mineral concentrations, can be considered extreme environments for life and provide ecological niches for highly specialized microorganisms. Dripping water, visitors and animals can provide organic input that facilitates life of heterotrophic microorganisms in some caves. Microorganism in caves is the main biological habitant and remarkably contributes to cave ecology.

Cave Vrelo is the deepest cave in Macedonia located in the canyon Matka which is home for many endemic species not found anywhere else in Europe. The aim of this work is to offer some preliminary data for microbiological diversity of sediments from cave Vrelo.

Samples were taken from 5 locations (depths: 14.5 m, 15 m, 47.5 m, 40 m and 100 m). These samples were taken in sterile containers and kept on ice until processing in laboratory.

All samples were investigated for several groups of bacteria, yeasts and molds using standard microbiological tools, while molecular identification of the isolated bacterial species was done by

sequencing of bacterial 16S ribosomal RNA gene. All of the investigated groups of microorganisms, except anaerobic sporogenic bacteria were present in sediment samples. Notably, a large number of coliformic bacteria (total and fecal) were isolated from all of the investigated samples which classify this water in Class IV, as ecologically unsuitable drinking water. Most of the identified non-coliformic bacteria belonged to the genus *Bacillus*. We have also identified representatives from *Staphylococcus*, *Proteus*, *Brevundimonas* and *Enterobacter*. Overall findings suggest a possible connection between the water from the cave and surface waters. Further investigation should be performed to determine the origin of these waters.

Keywords: microorganisms, cave, microbiological diversity, 16S ribosomal RNA gene.

Cyanobacteria and algae from biofilms: the comparison of phototrophic microorganism community from cave entrance and lampenflora - Lazareva cave, Serbia

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Phototrophic microorganisms inhabit the most various niches and many of them are known to grow and proliferate in habitats that in many ways account for extreme ones. This especially refers to cyanobacteria, microorganisms already known to have tolerance toward a wide range of environmental factors. Caves are also considered extreme environments, mainly because the low nutrient content that organisms use for their growth. However, the microorganisms' presence in caves is easily noticeable: spots on cave walls, discolorations, depositions and biofilms – complex communities of different phototrophic and heterotrophic microorganisms. Phototrophic microorganisms (cyanobacteria and algae) in caves are found at the places illuminated by natural light (entrances) and deep in the show caves around the artificial lights (lampenflora community).

We investigated and compared the entrance and the lampenflora phototrophic biofilm component from Lazareva cave, Serbia. Temperature (T), relative humidity (RH) and light intensity (LI) were measured and chlorophyll a (Chl a), water (WC), organic (OC) and inorganic matter content (IC) determined. Cyanobacteria and algae were identified according to the standard identification keys.

During the survey, Cyanobacteria, Chlorophyta and Bacillariophyta were documented. The entrance community was dominated by Cyanobacteria (with coccoid forms prevailing and *Chroococcus* and *Gleocapsa* species as the most common), while higher filamentous forms and those with heterocysts were less present. In contrary, the community around artificial lights was dominated by *Chlorella* sp. (Chlorophyta), while cyanobacteria appeared sporadically. *Chroococcus ercegovicii* and *Gleocapsa alpina* were present in both communities. Values of Chl a, WC, OC and IC, as well as physical parameters, also showed different trend at the entrance, than in cave interior.

Growth of microorganisms on cave walls and formations, especially lampenflora, is a serious

ecological and esthetic problem, because many of them produce metabolites that modify the rock surfaces and cause damage to these natural heritage sites.

Keywords: aerophytic cyanobacteria and algae, extreme habitats, conservation, Gloeocapsa, Chlorella

Diatoms diversity and ecological status of the Đetinja River (Serbia)

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The Đetinja River is located in western Serbia and it was dammed for creating the artificial Vrutci Lake. Samples were collected during 11 sampling events (September, October and December 2014, March, April, May, June, July, August, September and October 2015). Epilithic diatoms were scraped from the stones by brush. Permanent slides were observed by Zeiss AxioImagerM.1 microscope with DIC optics at magnification x1600 and AxioVision4.8 software. The relative abundance of diatoms was estimated by counting 400 valves of each taxa. The biological assessment of water quality was performed using diatom indices calculating by OMNIDIA 5.3 software.

Total of 113 diatom taxa have been recorded in the studied samples. *Achnanthydum minutissimum* was abundantly developed in mass and predominated over the other diatoms in almost all sampling periods. In some periods it was followed by *Amphora inariensis*, *A. pediculus*, *Cocconeis placentula* var. *lineata*, *Gomphonema olivaceum*, *Nitzschia amphibia* and *N. fonticola* as dominant or subdominant taxa in the community. The diatom indices analysis showed good to very good ecological status of the Đetinja River in the investigated period. They indicated absence of organic pollution. In almost all sampling events anthropogenic eutrophication was low, except in March when it was moderate. In March *Gomphonema olivaceum* was dominant taxon in the epilithic diatom community as β -mesosaprobous, eutraphentic taxon.

The Đetinja River was characterized by high species diversity and good to very good ecological status which matches with the literature data about rivers and streams with low level of pollution which are characterized by high species richness.

Keywords: *Achnanthydum minutissimum*, *Gomphonema olivaceum*, diatom indices, OMNIDIA.

Epilithic diatoms of Tara River (Montenegro)

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Biomonitoring tools for aquatic ecosystems are largely lacking for many developing countries, resulting in adoption of tools developed by other countries. Apart routine bacteriological analyses, the Montenegrin national monitoring system does not include biological parameters. The aim of this study was to estimate the use of epilithic diatoms in ecological assessment of the mountain rivers in Montenegro in order to contribute to the development of a widely applicable methodology for water-quality monitoring. This study represents the first diatom based water quality assessment of running waters in Montenegro carried out according to the recommendations of the Water Framework Directive. Water and epilithic diatom samples were collected from a typical mountain river (Tara River) and its main tributaries, in two seasons (spring and summer) in 2015. Investigated waters characterize high pH, alkalinity and oxygen concentration and low nutrients concentration. A total of 52 diatom taxa were identified and most of them, especially the dominant ones, were typical inhabitants of alkaline, nutrient-poor, fast-flowing waters of mountainous regions. Selected diatom indices (Trophic Diatom Index and Biological Diatom Index) indicated very good water quality (i.e. water quality class I) of the Tara River. High degree of compliance between diatom indices and water quality variables (e.g. nutrients) observed in this study, showed that selected diatom indices, if implemented in Montenegro, would provide a valuable addition to Montenegrin suite of tools for the biological monitoring of the water quality. On the basis of the coevaluation of biological and chemical parameters, Tara River seems to be the appropriate reference site for the classification of the highland alkaline rivers of Montenegro. However, further investigations, extended to other rivers in this region, are needed for the final development of the biomonitoring tools which would contribute to more comprehensive ecological assessment of Montenegrin rivers and enable harmonization with claims of WFD.

Keywords: diatom indices, ecological assessment, epilithon, water quality, Tara River, Montenegro

Macrozoobenthos of the Mediollitoral of Sazani Island (South-Eastern Adriatic Sea, Albania)

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Knowledge on the macrozoobenthos of Sazani Island is limited, because marine communities in this area have been very scarcely investigated in the past. This paper represents data on benthic macroinvertebrates from the rocky coast of the island that has been investigated during 2012 – 2014. Samplings have been carried out in four sites, of which two on the eastern coast and two others on the western coast of the island. Replicated quantitative samples have been taken in spring and in autumn

each year, by using a reticulated frame as a standard sampling area unit. This study gives data on species composition of macrozoobenthos and a general assessment of quantitative characteristics, seasonal variations and stability of macrozoobenthic populations in the studied area. A high predominance of mollusks has been recorded in the samples, followed by crustaceans, polychaetes, cnidarians, nematodes and sipunculids. It is worthy to note the presence of several benthic species, associations and habitats that have been considered of special importance in regional and international scale. The highest abundance has been recorded for gastropods and crustaceans. The seasonal variations were relatively high in both species number and abundance. The stability of benthic community seems to be moderate in most sampling sites. Comparing the eastern and western coasts of the island, there are evident differences in species composition, quantitative characteristics and degree of stability of the macrozoobenthic population between the two sides. Algal cover and exposure of the coast seem to play an important role for the species composition and abundance of benthic populations of the island. Urban and tourist developments in Vlova Bay have continuous impacts on the island and its benthic communities, especially on the eastern side of the island.

Keywords: benthic populations, rocky coast, habitats, sampling sites

Hidrochemical regime in the water of fishponds of PWE “Strezevo”

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One of the multipurpose activities of the reservoir “Strezevo” as one of the largest accumulations in Republic of Macedonia is fishery. The reservoir offers favourable conditions for the development of commercial fishing, but it is also favorable as well as for sport fishing tourism. Growing rainbow (Californian) trout in fisheries in the hydro system is an activity that has a great future and significant economic justification, because it is a high quality meat with a high degree of profitability.

Due to get higher quality profits of California trout or fish meat quality, the water quality in the fishfarm of HS Strezevo is continuously monitored. In the period from January 2014 to December 2014, collecting water samples from the entrance and exit of the pond (fishfarm) is made by monthly dynamic. Physico-chemical analysis of water from the pond covers the following parameters: temperature, dissolved oxygen, turbidity, conductivity, free carbon dioxide, ammonia nitrogen, nitrate nitrogen, pH.

The obtained results of the research showed that the pond water satisfies the required quality standards, which has realized the conditions for successful cultivation of rainbow (California) trout in the fishfarm (pond) which is within the HS Strezevo.

Keywords: Strezevo reservoir, fishpond, rainbow (California) trout

The impact of trout farm effluent on diatoms richness in the Rasina river (Serbia)

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The Rasina River is right tributary of the Zapadna Morava River, situated in central Serbia. The samples were collected in spring (April), summer (July) and autumn 2011, from 6 localities along the river at an altitudes between 640 – 704 m a.s.l. Sampling sites are located before and after trout pond. Epilithic samples were scraped from the surface of stones by brush. Light microscope observations and micrographs were made using a Zeiss AxioImagerM.1 microscope with DIC optics and AxioVision 4.8 software. At least 400 valves were counted in order to calculate relative abundance (%) of each taxon. The biological assessment of water quality was performed calculating diatom indices by OMNIDIA 5.3 software.

A total of 166 taxa belonging to 45 genera were identified in the studied samples. The most numerous were taxa of genus: *Nitzschia* (26), *Navicula* (22) and *Gomphonema* (15). The dominant taxa during spring were *Achnanthydium pyrenaicum* and *Diatoma ehrenbergii*, during summer were *A. pyrenaicum* and *Cocconeis placentula* var. *lineata* and during autumn was only *C. placentula* var. *lineata*. Diatom richness before trout farm was higher than after, which confirms its impact. The highest diversity was in spring and constantly decreasing through seasons.

The Rasina River was characterized by good to very good ecological status of water, with slightly differences between the values of the diatom indices during the seasons (spring, summer, autumn, respectively).

Fish feed remains can cause enrichment of the water with nutrients (in the form of ammonia, phosphorus, nitrogen) and most often leads to eutrophication which can be seen on the basis of the dominant species during the seasons. Change of dominant species between the seasons in the Rasina River was correlated with the values of nutrients.

Keywords: *Achnanthydium pyrenaicum*, *Diatoma ehrenbergii*, *Cocconeis placentula* var. *lineata*, diatom indices, nutrients.

The impact of trout fish ponds on the ecological status of the Vrla and Mlava rivers (Serbia) based on epilithic diatom communities

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Physical-chemical parameters of habitats are of great importance for the development of aquatic vegetation. Anthropogenic activities, such as the construction of fish ponds, can result in changes in environmental parameters. The Vrla River is right tributary of the South Morava River in the southeast of Serbia. The Mlava River is located in the eastern of Serbia, right tributary of the Danube River.

The main objective of this study was to determine influence of trout fish ponds on the ecological status of the Vrla and Mlava rivers based on the epilithic diatom communities.

The algological samples were collected from six sites in the Vrla River and five sites in the Mlava River during six seasons in 2011 and 2012. Standard method with concentrated sulfuric acid was used for diatom frustules cleaning. Relative abundance of each diatom taxa was determined by counting at least 400 valves on each slide. Seventeen diatom indices were calculated using the OMNIDIA software.

The water of the Vrla and Mlava rivers corresponded to high and good ecological status. Diatom indices calculation showed that on studied sites there are no major variations in water quality regardless of the trout fish ponds in both rivers. Although, some indices had the highest values at sites located upstream of the trout fish pond in the Vrla River. There is a possibility of organic pollution on sites located downstream of the trout fish pond in the same river. Values of diatom indices indicated high and good ecological status along the entire investigated section of the Mlava River, but, as well as in the Vrla River, the values of some indices were higher at sites located upstream of the trout fish pond. The obtained results show that the trout fish ponds don't affect significantly negatively on the ecological status of studied rivers.

Keywords: anthropogenic activities, diatom indices, environmental parameters

Influence of land-based trout farms on stonefly (Insecta: Plecoptera) larvae assemblage

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Activities related to agriculture are one of the major problems of freshwater pollution. Even though the influence of such practice is lowest on highland streams and rivers, fish farms localized in the upper course could disturb their homeostasis. Since trout farming has become an important economic activity in Serbia, three trout farms with different annual production were chosen in order to monitor their effect on sensitive stonefly larvae assemblage. The trout farms with low, medium and high annual production are situated along Studenica, Crnica and Mlava rivers, respectively. Research was conducted every two months during the one-year period. Water and biological samples were collected at one control locality and four localities downstream from the trout farms. Water temperature, pH, dissolved oxygen and conductivity were measured in situ, while phosphates and ammonia in the laboratory. Canonical Correspondence Analysis was conducted in order to test how taxa within the Plecoptera assemblage were grouped in relation to measured environmental parameters. Considering CCA analysis, the most important parameters that shaped the Plecoptera assemblage were phosphates and temperature. The greatest diversity of stonefly larvae assemblage was noted in the Studenica river, while the lowest was at the Mlava river, with its absence at the locality immediately downstream from the trout farm.

Taking into account that trout farms may be one of the major sources of pollution of highland streams, it is necessary to control and improve the trout production in order to protect overall stream community, especially the most sensitive ones.

Keywords: trout farming, bioindicators, water parameters, organic pollution, environment protection

With the olm (*Proteus anguinus*) we share dependence on clean groundwater

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The olm (*Proteus anguinus*) is the only cave-dwelling vertebrate of Europe. It inhabits subterranean rivers of the Dinaric Karst in the Western Balkans. *Proteus* is an endangered amphibian, protected by national legislation in Italy, Slovenia, Croatia, Bosnia and Herzegovina, listed on the Bern Convention and the EU Habitats Directive, and defined as vulnerable by the IUCN.

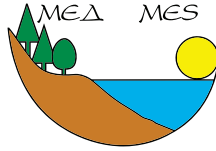
The Tular Cave is a natural cave in Kranj, Slovenia, turned into a laboratory in 1960 by speleobiologist Marko Aljančič. Tular is the only cave laboratory specialised on studying *Proteus*, its ecology and behaviour, and has an established *ex situ* breeding program. All experiments are based on observation, carefully designed not to harm or stress the animals. Since 2002, the black *Proteus* (*Proteus anguinus parkelj*) the most rare of all *Proteus* populations, discovered only 30 years ago in Southeast Slovenia, is also studied in the laboratory. The facility also serves as a sanctuary for injured *Proteus* accidentally washed out of their subterranean habitat during seasonal flooding.

Nowadays *Proteus* is threatened by pollution of groundwater and destruction of its subterranean habitat. Most threats came from intensive agriculture, hydroelectric energy production, and unregulated urbanization. Groundwater is the only reliable source of drinking water in karst areas, and diminishing *Proteus* populations indicate severe pollution.

Knowledge on the distribution of *Proteus* is urgently needed for an efficient protection and understanding of the species. Due to inaccessible subterranean habitat, its presence can only rarely be confirmed by classical survey methods. The Tular Cave Laboratory is introducing a method to detect traces of *Proteus* environmental DNA in groundwater. In 2015, utilizing the eDNA, new localities of the extremely rare black *Proteus* were discovered.

Proteus and its cave habitat represent one of the world's prime symbols of subterranean biodiversity, a flagship emphasizing the high vulnerability of European karst groundwater.

Keywords: human fish, pollution, karst, vulnerability, cave biodiversity



Section 5

FORESTRY AND AGRICULTURE



Multiyear program of work on animal genetic resources in Slovenia

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The vision on conserving of biodiversity for food and agriculture and promoting its use in support of global food security and sustainable development, for present and future generations. The Animal Genetic Resources for food and agriculture (AnGRFA) are a common concern of all countries, in that all countries depend on genetic resources for food and agriculture that originated elsewhere. The countries are obligate strives to halt the loss of genetic resources for food and agriculture, and to ensure world food security and sustainable development by promoting their conservation, sustainable use, including exchange and the fair and equitable sharing of the benefits arising from their use. The first Slovenian Multiyear Program of Work on AnGRFA was approved by Slovenian Government in period 2001-2009, the second Slovenian Multiyear Program of Work on Animal Genetic Resources for Food and Agriculture (MyPoW AnGRFA) was approved by Ministry of Agriculture, Forestry and Food MAFF) for period of 2010-2016. The Program exist all four mentioned Strategic Priority Areas. In the area "Policies, institutions and capacity building", the international cooperation exist great stress. The third period MyPoW was prepared for period 2017-2023, which follows the FAO Global Plan of Action for AnGRFA and Interlaken declaration. These mean, that MyPoW of Slovenia containing all 23 Strategic Priorities Area for Actions.

Keywords: food, agriculture, Animal Genetic Resources for food and agriculture, Slovenian Multiyear Program of Work

Genetic resources of agricultural crops in the region of river Bregalnica basin

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The agricultural production in Macedonia is consisted of cultivation of commercial varieties and hybrids on large areas and landraces on small fields and gardens for own use. Until today, the distribution and diversity of landraces is not evaluated scientifically. Regarding this, inventarization of the landraces only was carried out in the region of the river Bregalnica basin. Information and seed samples (3770 in total) were collected in 150 villages in 14 municipalities.

Among the cereals, highest number of samples with valuable diversity was collected for maize (335). Farmers maintain mainly old commercial varieties of wheat and barley, while oat and rye landraces are facing extinction. Most valuable diversity was noted for beans which are most distributed legumes with 680 samples of dry and 132 of green beans collected. According to the seed characteristics of dry beans only, more than 300 different landraces were identified. On the contrary, very few samples of chick pea, pea, lentil, faba and adzuki bean were collected indicating that their

maintenance is endangered. The highest diversity of vegetable landraces was identified in pepper (460), pumpkins (405), tomato (248), melon (114) and watermelon (88 samples). Few samples of other vegetables were registered, while onion and garlic are frequently distributed with insignificant diversity. Landraces of the industrial crops cotton, hemp, flax, sesame, peanuts that were cultivated 50 years ago are completely lost. Few samples of poppy and sunflower landraces were collected indicating their extinction in near future. Forage crops have similar status as their cultivation rely on commercial varieties only. All samples should be further characterized and evaluated experimentally in order to identify duplicates and accurate number of different landraces. This research confirmed that Macedonia has rich agribiodiversity and that significant part of it will disappear in several years. Therefore urgent activities for landraces conservation should be undertaken.

Keywords: agribiodiversity, landraces, seed samples, inventarization, extinction

Use of crop genetic resources in breeding programs for food security and sustainable agriculture

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During recent years, the awareness of the importance of agricultural biodiversity, conservation for sustainable utilization and improvement has been increasing. The world's genebanks hold bounties of diversity, mostly cultivars, landraces created by farmer-assisted evolution and wild relatives. Today, there are over 1700 genebanks in the world, conserving more than 1.5 million seed samples. Macedonia is considered as one of the world's biodiversity hot-spots. However, crop genetic resources have never been systematically collected, neither completely characterized or utilized. Moreover many of them are facing diversity loss. For that purpose, the Department of Genetics and Plant Breeding at the Faculty of Agricultural Sciences and Food in Skopje started a collecting mission in 2015 with the aim to preserve old landraces in Macedonia for long-term use. The established seed collection currently comprises 5523 samples of landraces only, belonging to 66 crops, collected from 429 sites in 69 municipalities. The preliminary analyses of information and seeds indicate presence of valuable genetic diversity within the collection, especially among legumes and vegetables. Several crops and numerous landraces are already lost, while many will be extinct in near future. In conclusion, there is an urgent need to collect seeds from the whole territory of the country, to characterize them and to develop core collections based on unique genetic diversity information. In that way, the available resources will be exploited in breeding programs and promoted for direct production as added-value cultivars.

Keywords: agricultural biodiversity, seed collections, genebanks, breeding, Macedonian landraces.

The impact of artificial wcr eggs infestation on stem diameter as morphological characteristic of maize

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The Western corn rootworm (WCR), *Diabrotica virgifera* sp. *virgifera* (Col., Chrysomelidae) is native in America. Represent economically one of the most important maize pest worldwide. After introduction in Europe 1992 in Serbia today is wide present in maize field thru the continent, living large damage on plant and yield.

Field experiment was carried out in 2014 and 2015 in Vojvodina province (loc. Bečej). The aim of study was to measure influence of root feeders on stem diameter of plant. The field chosen for the experiment represents a field with the low natural WCR infestation. During both experimental years, 96 maize plants were selected, labeled and arranged into pairs. The plants are set up in two rows with space between labeled plants of at least 1m. Each pair consisted of one plant artificially infested with WCR eggs (D) and the control plant (C). In D root zone injected 4 mL of WCR eggs 0.125% agar suspension (136 eggs/mL), and in C plant root zone 4mL of distilled water.

During the two years the plants in experimental field were inspected twice and measured stem diameter. Collected data statistically analysed by Kruskal Wallis one-way analysis of variance. Obtained results indicate statistical differences between C and D plants in both years. Analyses of stem diameter per year indicate statistical differences at the level of $P \leq 0.05$ between C and D plants. In the other hand significant statistical differences at the level of $P \leq 0.01$ registered inside plant groups per year. According to obtained results, high influence on biology, population dynamic and harmfulness of pest realize agro meteorological factors i.e. temperature and precipitation. These factors highly influence number of root feeder's larvae in the soil and level of attack on the root, and in that way respond to stem diameter.

Keywords: *Diabrotica virgifera*, influence, corn, thickness

Estimation of effective size of Busha cattle in the R. of Macedonia

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Busha cattle is an autochthonous breed in the R. of Macedonia. It belongs to a group of primitive shorthorn cattle (*Bos brachyceros europaeus*). In Macedonia this breed has officially been classified as triple purpose breed (for meat, milk and work) but considering its low productive capabilities it is more similar to some primitive working breeds. This breed of cattle used to be dominant and most important breed in almost all Balkan countries until 50s and 60s of the XX century but today in lowland areas

with intensive farming they are already replaced with more productive and specialized cattle breeds. Today these cattle are no longer used for work but because of absence of systematic cattle improvement program these animals have retained their poor milk and meat production capability. This breed is well adapted to the very harsh feeding and housing conditions that exist in the rural areas of the Macedonian mountains and is resistant to diseases. It is still the most significant milk and meat resource for these areas where the more productive cattle breeds cannot thrive successfully. The effective size of busha cattle in the R. of Macedonia in 2015 was 239.80 and according to the total number, this population is in risk. Also, the present problem is to get enough quality selected Busha bulls aimed for reproduction.

Keywords: Busha, cattle, domestic breed, effective size.

Variation of milk yield and milk composition of the autochthonous Ovchepolian sheep breed

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The Ovchepolian pramenka is a native sheep breed in the country and it is included in separate sheep selection breeding program. The aim of this research is to present the results on the milk traits and variation of milk composition during lactation of Ovchepolian pramenka breed. Milk recording was performed according to the ICAR regulation and milk yield was evaluated by the AT4 method. The following milk traits were analyzed: length of lactation, milk yield (daily milk yield, milking yield and total milk yield) and milk composition (fat and protein content). A total of 450 completed lactations were analyzed in a period of three years (2013, 2014 and 2015). Average length of lactation was 155, 153 and 156 days, with an average milking yield of 67,671 (65,32 kg), 68,231 (65,86 kg) and 83,541 (70,61 kg) in 2013, 2014 and 2015 respectively. Differences in milk composition among lactations and test days (days of milk recording) during the lactation period were significant ($P < 0.01$). The average milk fat content in 2013, 2014 and 2015 was 7,56%, 7,65% and 6,16% respectively, while the average protein content was 6,47%, 6,39% and 6,25% respectively. Achieved results suggest that a more detailed genome selection programme of the Ovchepolian sheep breed, encompassing udder selection as well as molecular characterization, should be applied.

Keywords: lactation, pramenka, fat, protein, selection, native.

Climate changes and adaptation capacity of autochthonous poultry genotypes

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Growing human population forces agriculture to produce more valuable proteins, challenging this goal of increased production with sustaining or improving the environment. Climate change, food security, ecology impose a shift toward monogastrics and breeds that are efficient converters of feed into meat, milk and eggs. Evaluation of the livestock species' and breeds' vulnerability to climate change is explored through the systematic searches for adaptation traits in local, indigenous breed's diversity.

Climate changes mostly related with increased temperatures affects existing production systems. Chickens have limited capacity of thermoregulation above 30°C, and the result is drop of egg production and egg quality. Genetic diversity and adaptation potential of local breeds has been mentioned as essential value in future changes in production conditions and therefore different scenarios of climate change mitigation strategies stress that local genotypes should be protected, conserved and secured. Is there real "genotype" possessing adaptation traits in local breed and populations, or it are the "each indigenous locally adapted breeds" that interacts with their habitat, feed and climate conditions especially temperature to forms the "complex of adaptation traits"? Therefore better characterization of breeds their genetics and their expressions and interactions in different production environments needs to be constantly evaluated.

Production and egg quality parameters were followed in a flock of autochthonous chickens in R. Macedonia before and after the onset of high summer temperatures (May, June and July, August) to explore if the heat stress effect of high summer temperatures has negative influence on this genotype.

Production data (number of the eggs – egg laying percentage) and egg quality parameters (and mass-size, eggshell strength, yolk colour, albumen quality-Hough units) followed, show drop of all these parameters under heat stress conditions (at the onset of season of high summer temperatures). Significant drop in laying percentage (80.19 before and 70.32% during the high temperature) egg size (59.85g vs 49.43g), eggshell strength (3684.37 vs 2987.67g/cm²), and yolk colour (12.50 vs 9.88 Roche units) but not for albumen quality (60.12 vs 61.55 Hough units), were monitored before and during the heat stress, respectively.

Local, indigenous genotype of chicken reacts on heat stress with drop of production and egg quality parameters. It is not entirely clear if this drop can be associated only to the heat stress or other factors (feed availability and quality, housing or management) contribute to it. Based on these arguments, statements that local genotypes are more adaptive to poor environmental conditions and harsh feed has to be reinvestigated and their adaptive potential to combat increase in temperatures (heat stress) should be monitored in interaction of genotypes and other factors like feed availability, housing and management conditions in total.

Keywords: indigenous poultry, environment, heat stress, productivity

Productivity and sustainability of autochthonous Macedonian poultry genotypes

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Variety of phenotypes and genotypes exist in the flock of chickens in the rural areas in R. Macedonia. Presumption was that the original, descent autochthonous genotype is preserved somewhere in these rural areas flocks. After thorough check, only small number of individuals with genotypes resembles descent autochthonous genotype, were phenotypically characterized as indigenous chickens. Phenotypic characterisation defines several possible plumage phenotypes among these autochthonous genotypes village chickens. Egg production data records on these chickens are lacking due to extensive outdoor, backyard keeping (free range). Numerous, various, different nutrition and environmental conditions contribute "complexity factor" toward difficulties in evaluation of the production capacity of these birds, but according the breeders information they lay 100-120 eggs/year. Part year (24weeks; June-November) egg production recording was performed, after spring breeding season in three flocks separated by plumage colour phenotypes (ashy-silvery-brown leghorn laced, black with "Columbian" red on the neck, and grey or "blue") as a base for evaluation of the productivity level and estimate the model for economic sustainability of these autochthonous genotypes in comparison with the modern hybrid layers. Model is developed on the base of lost of income due to lower egg production. Ashy-silvery-brown leghorn laced variety produce 112.14, black 115.33 and "blue" 112.6 eggs/168days recording period. Recorded average laying percentage of 67.48% was base for estimating productivity level of 179eggs/year, presuming that only 265 days of production period exist and 100 winter days are empty production days due to harsh feeding and housing condition. Economic sustainability index $(310-179=131 \times 5=657)$ is estimated on the level of 657denar based on the income lost formula ((hybrid genotype yearly number of egg produced – autochthonous genotype yearly egg produced) x average egg price-5denar). Programmed state financial support on the level of 11Eur or double price level of "free range eggs" produced from these autochthonous genotypes should be adopted to compensate the loss of income and ensure sustainability in breeding these chickens in original condition of the rural households.

Keywords: indigenous poultry, productivity, sustainability

Ethological and production traits of native honey bee subspecies (*Apis mellifera macedonica*) in Republic of Macedonia

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In 2015 in honey bee colonies (n=100) from four genotypes (A, B, C and D) of the native honey bee subspecies *Apis mellifera macedonica* breed in Ohrid region, ethological and production traits were observed. The research included: scoring of defensive and swarming behaviour of honey bee colonies-gradation 1 to 4 according to Ruttner method, 1972; scoring of the hygienic behaviour of honey bees using Pin test method as well as scoring of the honey productivity by weighing of extracted honey in kg per bee colony.

The results did not show statistically significant differences concerning defensive behaviour between colonies from the four genotypes which, by the average scoring for defensive behavior (A-2.84, B-3.0, C-3.04 and D-2.8), may be characterized as moderately defensive, but with increased mobility and sensitivity.

Related to swarming behavior, for which scoring values are ranged from 3.92-4.0, colonies from all genotypes were positively scored, because they did not manifested expressive swarming behavior, and statistically significant differences between bee colonies were not identified.

Significant statistical difference between researched bee colonies was indicated in regard to the hygienic behaviour. Genotype A is scored as line with most expressive hygienic behaviour, until genotype D as line with least expressive hygienic behaviour. Yet, statistical significant differences were determined between A and D ($p < 0.05$), and between B and D ($p < 0.05$) lines, which gives opportunity for choosing queens for selection purposes.

The average honey yield per colony in the researched genetic lines ranges between 10.9 and 11.6 kg per bee colony, which is very close to the national average in the last ten years (www.stat.gov.mk 2016). There was also no significant statistical difference concerning this characteristic.

Keywords: *Apis mellifera macedonica*, swarming behaviour, defensive behaviour, hygienic behaviour, honey production.

The essential role of traditional agriculturist in landscape modelling: Example of Croatia

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There are four basic types of environment in Croatia - lowland, highland, Mediterranean with the islands and the Adriatic Sea which consist of sixteen basic landscape types with distinctive visual

identity and diverse habitats. The majority of Croatia's landscapes are semi-natural, shaped over a long time period of human and nature's coexistence. Traditional agriculture and livestock keeping activities turned vast areas of wetlands, mountain forests and karst fields into open landscapes abundant in diversity of habitats such as the rural landscapes of Lonjsko polje, Žumberak, central Istria, Dinaric karst fields of Lika, Neretva valley, etc. In these areas anthropogenic influence can be seen in the plough fields, orchards, vineyards, olive groves, pastures, meadows, dry stone walls, ponds, wells, pens, stables, as well as in the effect on the relief, particularly on vegetation and water. In the need for fertile land, karst areas were cleared of rocks, which were then used to build *suhozidi* (dry stone walls) and *dolci* (small depressions), *torovi* (livestock huts), *stanovi* (farmer/shepherd huts), *bunje* (field huts), *lokve* (ponds) and *bunari* (wells), all of which are distinctive elements of the karst landscape, some created already during the Neolithic age. Some traditional agricultural landscapes have an outstanding value such as the Starigrad polje on the island of Hvar - the best preserved Greek system of agricultural land division on the Mediterranean, included in the UNESCO World Heritage List. Landscapes have mainly not yet been evaluated in the sense of their ecological value and economic use even though they have an exceptional potential for local development based on organic agriculture, cultural heritage and tourism. Their protection should include all activities which ensure preservation of their features, primarily traditional agriculture through which they were created. The protection of Croatian landscapes and the accompanying biodiversity therefore strongly depends on the traditional agriculturist.

Keywords: agriculture, agrobiodiversity, traditional architecture, protection, valorization

The Green book of Croatian breeds: a concept of endangerment assessment in agrobiodiversity

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Indigenous cultivars, plant varieties and animal breeds, are an important part of biodiversity as well as of cultural heritage. Recognizing their importance in landscapes, ecosystems, associated habitats, and connected biodiversity is obligate duty, especially in Balkan region, recognized as one of Europe's biodiversity Centers. During 20th Century, industrialization of agriculture caused decrease of agrobiodiversity and separation of agriculture from natural ecosystems, followed by degradation and disappearance of some agro-ecosystems, agro-habitats and many traditional cultivars. Experts team, supported by the Croatian State Institute for Nature Protection (SINP), have developed a Green book concept, methodologically based on a concept of IUCN Red books, but also combining FAO, EU and National program for preserving domestic animals breeds, resulting with basic estimation of endangerment, but also protection and conservation directions. In total 12 species of domestic animals are recognized, belong to 2 phyla, 3 class, 6 orders and 7 families, with 62 traditional breeds: 10 sheep; 8 pigs, chickens and dogs; 7 horse and cattle; 3 donkeys, goats, geese and turkey and one for ducks and bees. Through Green book many aspects were analyzed: Croatian/English name; synonymy; threat category (FAO/EU/IUCN/NKU); origin; cultural heritage value; ecology; description; population trend and distribution; current value; endangerment causes; current regulatory protection; conservation measures. According to methodology, 12 breeds are probably extinct (?EX), 9 globally and 3 regionally;

3 critically endangered (CR); 13 endangered (EN); 12 vulnerable (VU) and 5 not threatened (LC). For 16, status is not established due to data deficiency (DD), which indicates necessity of further researches, supported by only 26 officially recognized breeds versus 62 found. Agrobiodiversity analyses should amplify on other cultures, but also conducted on Balkan region, with establishment of protection and preservation measures and evaluation of economical use for autochthonous breeds.

Keywords: agriculture, farming, animal husbandry, cultivars, vulnerability, protection

The importance of indigenous breeds of horses in cultural heritage, landscape and biodiversity protection and sustainable models of their conservation

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Over the history horses have lost their primal use which was related to agricultural labour, transport of people and goods and warfare which has, in addition to technological development, caused a strong decline in their number. Throughout this long history numerous breeds of horses were shaped to suit human needs, specific habitat conditions and local cultures. Since the usage and husbandry of horses today is mainly related to sports and recreation, indigenous breeds hardly find a space for survival. Their significance and role in landscape maintenance and biodiversity conservation is on the other hand undisputable due to their specific adaptations to local ecosystems. Traditional breeding practices on wetlands, natural and semi-natural grasslands are a necessity for conservation of these ecosystems. Role of these breeds in traditional customs such as „Sinjska alka“ is an irreplaceable part of local cultural identity. It is therefore necessary to develop new models of horse usage which are based on traditional breeding practices and preservation of indigenous breeds' genotypes. In order to be sustainable these models should be economically, socially and environmentally feasible. These requirements can be reached if husbandry and usage of these breeds are associated with tourism, human health and nature protection. Such models of sustainable breeding practices of the indigenous breeds Croatian Posavina horse, Lipizzan and the younger breed – Croatian warm-blooded horse, have been analysed and their justification has been proven creating added value in local economies of rural areas and ensuring stability of horse population in the environment. A horse breeding farm oriented towards milk production in Austria has been analysed and presented as a good practice example of the indigenous horse breed conservation through new models of usage. Followed by good organisation, business plan, and marketing strategy, given model can easily be implemented in countries such as Croatia or Macedonia.

Keywords: horse breeds, adaptations, genotypes, traditional agriculture, milk production, ecosystems

The White Book: Traditional sorts and breeds of Dalmatia: a model for agrobiodiversity inventory for a whole Balkan region

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Dalmatia is a historical name for a Mediterranean region of coastal part of SE Croatia. High value of agrobiodiversity, Dalmatia thanks to the high diversity of landscape, climate, pedosphere and general biodiversity, as well as its position in Mediterranean basin, Balkan peninsula and Dinaric chain, but also stormy political, cultural and socio-economic factors during history. Unfortunately, agrobiodiversity of Dalmatia is disappearing very quickly due to depopulation, deagrization and industrialization before it has been sufficiently researched, preserved, protected and used. Agricultural biodiversity of Dalmatia has been recognized in frame of COAST project (2006 - 2013) conducted by the United Nations Development Programme (UNDP), in cooperation with the Ministry of Environment and Nature Protection. The aim of the project was to promote the Green Rural Development programme in Dalmatia based on local resources, traditions and environment, and implemented with the financial support from the Global Environment Facility (GEF). Traditional sorts and breeds of Dalmatia, as part of general biodiversity, with creation of database and published book, include the total of 526 varieties: olives (42); grape (83); other fruits (110); vegetables (41); arable crops (113); aromatic plants, herbs, healthy and honey plants, ornamental and wild edible plants (106) and domestic animals (31). This agrobiodiversity is constituted by high percentage of autochthones traditional cultivars, especially vineyards with 82 varieties, olives with 39 and other fruits with even 94 varieties, 30 breeds of domestic animals and high number of wild species with high agro-potential. Identification, inventory and preservation of agrobiodiversity is a key for creation of original, premium agriculture products, development of touristic offers and future plant selection. It represents an important tool for conservation of beauty and value of present landscapes of Dalmatia, but also a model for agrobiodiversity inventory for the whole Balkan region.

Keywords: agriculture, traditional cultivars, UNDP, vulnerability, protection, development

Erosion intensity as a consequence of the natural and anthropo-zoogenic factors, case study: catchment of the Spilje-Debar reservoir

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Erosion phenomena and processes in the catchment areas and the torrential activity in riverbeds cause endangerment and destruction of the hydropower and hydro-ameliorative facilities and systems, settlements, roads, water supply and industrial facilities and systems, large areas of productive lands, threatening, degradation and destruction of the biodiversity and the environment in general.

In the distant past phenomena and processes of erosion, especially their intensity / potential

dominantly were the result of natural factors. Nowadays, when not accessible areas became available to man and his activities, therefore the erosion phenomena and processes and especially the intensity of erosion became more pronounced.

In this paper it is stressed the role and influence of the human factor in the spatial distribution of the phenomena and processes of erosion, with the main emphasis on the intensity of erosion. The analyses were made with analytical methodology for quantitative and qualitative assessment of phenomena, processes and erosion intensity. The analyses were done using the EPM methodology of prof. S. Gavrilovic with small modifications. Erosion intensity / potential was defined with analytical approach and direct field mapping of the erosion factors and the erosion coefficient using modern GIS techniques.

The result show that the most vulnerable areas from erosion and surfaces with extremely high erosion coefficient / intensity is concentrated in the immediate catchment area of the reservoir. These are areas with extreme unfavorable natural conditions / erosion factors (geology, soil and orography) and these are areas exposed to strong human impact.

Keywords: erosion phenomena and processes, erosion intensity, EPM, Siplje reservoir

Defining the erosion potential through methodologies based on analytical approach and measurements

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The erosion map of R. Macedonia has been made using the methodology of prof. S. Gavrilovic, with small adjustments and changes in certain segments, according to the specifics of the factors of erosion in our country. Simultaneously with the drafting of erosion map of the RM were conducted measurements of the deposited erosive sediment in almost all major reservoirs in Macedonia up till 1994. Through the measurements of the deposited sediment in the reservoirs the methodology of Gavrilovic (EPM) was confirmed for accuracy and calibrated accordingly.

The catchment area of the reservoir "Spilje-Debar Lake" is characterized with large diversity in terms of all natural conditions and factors and pronounced human impact in the immediate catchment of the reservoir. The erosion intensity / potential was defined with analytical approach and direct field mapping of the erosion factors and the erosion coefficient using modern GIS techniques.

In parallel with the previous stage also it was done bathymetric measurement of the reservoir in order to determine the amount of deposited erosive sediment in the reservoir from its formation until 2014. With information obtained from the measurements and the EPM, there have been defined vulnerable zones / areas of the reservoir.

In this paper it is presented the application of two different approaches, methodologies, whose outcomes are very identical, which confirms the conclusion that the EPM method of Gavrilovic can, and should be widely used in defining the conditions and criteria for the construction of new hydropower plants.

Keywords: erosion, erosion intensity, erosion potential, EPM

Validating the FAO Aqua-Crop model for irrigated and water deficient sugar beet

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Simulation models that quantify the effects of water on yield at the farm level are valuable tools in water and irrigation management. To address this need, FAO has developed a yield-response to water model, named aqua-crop model, which simulates attainable yields of the major field and vegetable crops cultivated worldwide. Although the model is simple, it gives particular attention to the fundamental processes involved in crop productivity and in the responses to water, from a physiological and agronomic background perspective. The ease of use of the model, the low requirement of input parameters, and its sufficient degree of simulation accuracy make it a valuable tool for estimating crop productivity under rain fed conditions, supplementary and deficit irrigation, and on-farm water management strategies for improving the efficiency of water use in agriculture. In this study, aqua-crop model is parameterized and tested for sugar beet under full (100%) and deficit (75, and 50% of full) irrigation regimes. The model is able to simulate the crop water use under very high *ET* and wind conditions. Furthermore, the model performed satisfactorily for the growth of aboveground biomass, grain yield, and canopy cover in the non-water-stress treatments and mild stress conditions, but it is less satisfactory in simulating severe water-stress treatments, especially when stress occurred during senescence.

Keywords: AquaCrop Model, canopy cover, biomass accumulation, grain yield, efficiency of water use.

Current state of agri-environmental indicators of Republic of Macedonia

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Agri-environmental indicators are a useful tool for analysis of the connection between agriculture and environment and identifying developments in this intensive interaction. Such indicators should consequently help to accomplish a better understanding of the complex issues in the field of agriculture and environment, to show developments over time, and to provide quantitative information.

In order to help the improvement of environmental performance of agriculture, Organisation for Economic Co-operation and Development (OECD) has established a set of agri-environmental indicators, developed in co-operation with Eurostat and Food and Agriculture Organisation (FAO). These indicators inform policy makers and society on the state and trends in agri-environmental conditions, and can provide a valuable aid to policy analysis.

Currently, there is a deficiency of continuous monitoring of indicators connected to the impact of agriculture on the ecosystems and environment in Republic Macedonia. A development and

continuous monitoring of applicable agri-environmental indicators is a key of valuation of the impact of agriculture on environment. Therefore, this paper will give an overview of current data and state of agri-environmental indicators in Republic of Macedonia and accordingly an analysis and evaluation of the sustainable development in the country.

Keywords: agri-environmental indicators, agriculture, environment, agricultural impact, agricultural monitoring

The role of plant biotechnology methods in sustainable agriculture

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Plant biotechnology is set of different scientific approaches and methods that are utilized to improve and modify plants for human and environmental benefit. Plant biotechnology can be used to meet the increasing need for food by improving yields, improving the nutritional quality of crops and recuing the impact on the environment. Plant biotechnology can assist to creation of varieties resistant to frost, droughts and floods, pests and disease, and other abiotic and biotic stresses. Similarly, development of plant biotechnology methods is a reach source of possibilities for creation of new agricultural genotypes, thus enriching the agricultural biodiversity.

This paper presents several *in vitro* methods with successful application results and particular concern for improvement of the biodiversity of horticultural crops, important for Republic of Macedonia. Utilization of the benefits of plant biotechnology will bring “economically sustainable” and “environmentally sound” agricultural production that shall be “socially equal”. It is a straight contribution of plant biotechnology to the sustainable agriculture.

Keywords: plant biotechnology, sustainability, agriculture, in vitro methods, horticultural crops

In situ and *Ex situ* gene conservation of domestic animals in the Republic of Macedonia

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The breed structure, population trend and size of native sheep, goat and cattle breeds in the Republic of Macedonia requires further evaluation, inventarization and characterization in order to preserve and develop proper livestock biodiversity conservation strategies. Conservation of animal

genetic resources requires appropriate maintenance of the biological diversity, but also sustainable utilisation as a base for future needs in animal breeding. National legislative framework relevant to conservation of animal genetic resources include: law for livestock breeding and seven regulations related to conservation of animal genetic resources, the most important being the national programme for conservation of livestock biodiversity 2011-2017. As result of complete utilization of *in-situ in vivo* and *ex-situ in vitro* methods of conservation of animal genetic resources in the period between 2011 and 2016, satisfactory results were achieved in autochthonous sheep, goat and cattle breeds. In the Ovchepolian sheep breed, where breeding organization was recognized with 7 flocks and 3511 heads (3058 breeding females and 453 breeding males), the gene bank accommodates: 6676 semen straws from 9 donors animals, 500 blood samples, and 25 frozen oocytes from 4 donor animals. In the Karakachanian sheep breed the gene bank accommodates: 204 semen straws from seven donor animals and 50 blood samples, unfortunately with no recognized breeding organization. In the Domestic Balkan Goat breed, where breeding organization was recognized with 10 flocks and 2505 animals (2149 breeding females and 356 breeding males), the gene bank accommodates: 569 semen straws from 21 donors animals and 350 blood samples. In the Busha cattle breed, where breeding organization was recognized with 42 members and 720 heads (654 breeding females and 66 breeding males), the gene bank accommodates: 1003 semen straws from 6 donors animals, 350 blood samples. Further activities should provide higher number of donor animals per breed, as well as successful conservation of embryos and oocytes.

Keywords: ovchepolian, karakachanian, goat, busha, animal genetic resources.

Adding value to Macedonian opium poppy landraces: Improvement of yield traits and alkaloid content

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Opium poppy (*Papaver somniferum*), is an economic source of morphinane alkaloids such as morphine, codeine, thebaine, narcotine and papaverine exploited by the pharmaceutical industry. With regard to increasing demand for these compounds on global and national market, breeding program for poppy was enforced. The aim of this study is to present an overview of the successfully applied conventional breeding for enhancing the yield and alkaloid content in opium poppy. A total of 127 poppy genotypes with different origin were evaluated during 2010 to 2015, on experimental field near Skopje. In the prebreeding germplasm consisted of 60 genotypes from different countries and 30 Macedonian landraces in 2010, 37 new landraces and selected breeding lines were subsequently added in the following years. Agromorphological and productive traits, as well as alkaloids content were observed every year and standardized on a year basis according to the cultivar Alkaloid1. The classification of the genotypes was done based on Gower distance and the dendrogram was constructed using UPGMA method. In order to identify genotypes with desirable traits two way cluster analysis was applied. Significant diversity of evaluated traits has been recognized in agro-climatic conditions

of Macedonia and genotypes were effectively classified according to all analyzed traits. The results enabled clear overview of the phenotypic diversity of studied germplasm. Moreover, it was identified that landraces originating from Macedonia expressed the best values for all desirable characteristics. Several advanced lines as new cultivar candidates with significantly improved seed and capsule yield as well as morphine and thebaine content were selected. They will be further exploited in the breeding program for hybridization and for direct selection of positive lines.

Keywords: poppy, germplasm, landraces, breeding, yield, alkaloids, breeding

Effect of ecological fertilizer Megagreen on some morphological and productive properties of rice (*Oryza sativa* L.)

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In this paper are presented results from the study of ecological mineral foliar fertilizer Megagreen in rice. Two treatments were studied- standard fertilization (control) and Megagreen treatment, in 5 varieties (*Kiziltan*, *Gala*, *Halilbey*, *San Andrea* and *Prima riska*), in field trial set up in Zade design. Basic fertilizer of 500 kg/ha NPK (16:16:16), and second split of 150 kg/ha Urea 46 % N were applied. In Megagreen treatment the seed was presoaked for 24 hours prior to sawing in 0.5 % Megagreen solution, and the crop was foliar treated twice during vegetation with equal concentration. Results were analyzed with ANOVA and LSD test.

The results show that the highest stem was measured in the control, in variety *Prima Riska* (91.40 cm), while the lowest in Megagreen treatment in *Kiziltan* (48.90 cm). Longest panicle was measured in Megagreen treatment in *Prima Riska* (18.80 cm), while shortest in the control in *Gala* (12.03 cm).

Maximum and minimum number of productive tillers/ m² was produced in the control, by variety *Gala* (528.33), and *Prima riska* (372.00) accordingly. The 1,000 kernel weight ranged from 42.00 g in the control in *Prima Riska* (the highest), to 31.22 g in the control in *Kiziltan* (the lowest). Highest biological yield was obtained in Megagreen in *Halilbey* (24,900.00 kg/ha), while lowest in control- *San Andrea* (16,850.00 kg/ha).

Maximum paddy yield was achieved in Megagreen in *Halilbey* (9,507.34 kg/ha), while minimum in Megagreen- *Kiziltan* (7,077.38 kg/ha). Highest head rice yield was measured in control in *Prima riska* (62.33 %), while lowest in control- *San Andrea* (52.07 %). Highest white rice yield was determined in the control in *Prima riska* (5,658.56 kg/ha), while lowest in Megagreen in *Kiziltan* (4,170.70 kg/ha).

Megagreen had significant positive effect on stem height, panicle length and head rice yield, but not on white rice yield.

Keywords: variety, foliar, stem, panicle, white, yield.

Effect of ecological fertilizers Herbagreen and Megagreen on some morphological and productive properties of rice (*Oryza sativa* L.)

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In this paper are presented results from the study of ecological mineral foliar fertilizer Megagreen in rice. Two treatments were studied- standard fertilization (control) and Megagreen treatment, in 5 varieties (*Kiziltan*, *Gala*, *Halilbey*, *San Andrea* and *Prima riska*), in field trial set up in Zade design. Basic fertilizer of 500 kg/ha NPK (16:16:16), and second split of 150 kg/ha Urea 46 % N were applied. In Megagreen treatment the seed was presoaked for 24 hours prior to sawing in 0.5 % Megagreen solution, and the crop was foliar treated twice during vegetation with equal concentration. Results were analyzed with ANOVA and LSD test.

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Megagreen had significant positive effect on stem height, panicle length and head rice yield, but not on white rice yield.

Keywords: variety, foliar, stem, panicle, white, yield.

Research on the development and structure of the crown of old-growth beech trees on Maleshevski Planini Mts.

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This research is focused on the elements that characterize the crowns of old-growth beech trees, which grow and develop in pure beech forests on Maleshevski Planini. Old-growth trees of Maleshevski Planini can be found individually or in small groups, but they are very important for biodiversity conservation in the forest as well as the preservation of natural and autochthonous ambience. Research is conducted through direct field surveys collecting data on the development and structure of tree crowns.

Furthermore, the data are processed through statistical methods with appropriate analysis and the obtained results are shown in tables and graphics.

The results obtained from the research indicate that the old-growth beech trees have strongly developed crowns, high vertical and horizontal elongation and a significant share of the crown in the total height of the tree. With increasing age they increase the crown elements that affect its projection, its shape as well as the ability to use the space for growth. Thus the development and structure of the crown of old-growth trees as an essential element of the tree, is directly dependent and construction and other parts of the tree as a whole. Highly developed canopy the old-growth trees enable the favorable conditions for their growth and survival because some have achieved age greater than 400 years, but does retain a dominant position in the forest stand.

Preserving these old-growth trees is of great importance to science because they are adapted to all environmental changes for a long time and have developed a particular genotype and phenotype which whom they survived, and had a big impact on the development and structure of the crown.

Keywords: treecrown, beech, old-growth trees, Maleshevski Planini.

Vegetation succession on the mountain pastures in the region of Malesh

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The research focuses on vegetation succession at mountain pastures by setting up representative sample plots of 500 m² and collecting data on the species, the number of individuals, their height, the diameter of the crown and the diameter at breast height, in order to determine the presence of species, their coverage and vertical structure in the trial areas.

The analysis of the vegetation succession on the mountain pastures in the region of Malesh was done according to standard methods of vegetation studies, which include: consulting the existing literature and documentation, collection of field data by setting up representative sample areas, analysis of collected field data, extrapolate and recommendations for further research and action for reclamation of mountain pastures in the region of Malesh.

The results indicate a clear link between declining human population and livestock and vegetation dispersion on the mountain pastures. At the pastures abandoned up to 10 years the coverage is 0.53; at the pastures abandoned from 10 to 20 years is 0.85; at the pastures abandoned from 20 to 30 years is 0.9 and at pastures abandoned more than 30 years is about 1,0. Patterns of vertical structure clearly demonstrate the dynamics of succession processes.

Based on the conducted research is determined that the process of livestock reduction, reduction of land use of mountain pastures and absence of silvicultural measures for their maintenance and rehabilitation leading to their overgrowth with forest vegetation, which in 30 years has ability to completely grown over and thus cause changes in the landscape and biodiversity of the area.

Keywords: Vegetation succession, region of Malesh, mountain pastures, vertical structure.

Assessment of silvicultural characteristics affecting the conservation status of Grecian juniper (*Juniperus excelsa* Bieb.) woods in Greek Prespa

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Greek juniper (*Juniperus excelsa* Bieb.) is found from Albania to Western India and from the Arabian Peninsula (Oman) to Kyrgyzstan and Ukraine. In Greece, it is found in Western Macedonia, at Nestos Gorge and in various locations of Eastern Rhodope and on the islands of Thasos, Samos and Evia. At Greek Prespes, the forests present irregular structure in various succession phases, leading to the dominance of broadleaved species and irreversible degradation of the priority habitat type “Endemic forests with *Juniperus* spp”, unless restoration efforts take place. The present situation of the habitat type is the combined result of the abandonment of grazing and woodcutting of the broadleaved species and the cessation of pollarding and woodcutting of Juniper trees for fisheries and constructions. In order to implement restoration measures for the improvement of the conservation status of juniper forests, the target structural features have to be described. Initially we distinguishing the vegetation types a) Juniperetum excelsae-foetidissimae (dense canopy juniper stands), b) Mixed, open canopy young forest, c) *Quercus trojanae*-Juniperetum excelsae (dense canopy mixed stands) and d) grasslands with sparse juniper trees. Then after fieldwork and bibliographical research we concluded that Juniperetum excelsae-foetidissimae is the most representative of the habitat type in terms of diagnostic species with the following silvicultural characteristics: a) canopy closure near 100%, b) around 215 trees/ha and c) basal area greater than 52m²/ha. The actual corresponding maximum values found at Greek Prespes and for a limited area were 56%, 280 trees/ha and 37.12 m²/ha respectively.

Keywords: Greek juniper, *Juniperus excelsa*, Prespa, restoration measures

Impact of the forestry management practices on montane beech forests on Deshat Mt. (Republic of Macedonia) on the down dead wood biomass and carbon quantity

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Knowledge on the carbon cycling and its binding in forest ecosystems is invaluable for understanding of climate change and consequences on the management of forest resources.

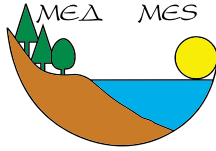
Results for the amount of down dead wood biomass (dead branches and dead trees) and content of carbon in five montane beech forest stands are presented in this paper. The investigation was conducted on Deshat Mountain within the Mavrovo National Park. The investigated stands represented five different types of forest in terms of their degradation and forestry practices (from the most preserved

forest with old and thick trees and large numbers of fallen trees where the forest biomass was not used for a long time, to degraded forest represented by a resprouting trees and a small amount of coarse biomass.

The results were obtained by two methods: a method of line transect for large branches (3-5, 5-10 and 10-20 cm) with test surfaces of 1m² and with assessment of biomass of all fallen trees on a specific test area. The down dead wood biomass (branches and fallen trees) was divided into four categories according to the degree decomposition. Carbon content was determined by the method of Kotzman.

The highest amount of down dead wood was estimated in the most preserved forest (19.04 t·ha⁻¹), and the lowest in the most degraded forest (2.68 t·ha⁻¹). Biomass of large branches ranged between 1.03 and 9.73 t·ha⁻¹, while the biomass of fallen trees varied between 1.65 and 11.64 t·ha⁻¹. The content of carbon follows the same patterns (10,99 t·ha⁻¹ in the most preserved forest and the lowest value of 1.54 t·ha⁻¹ in the degraded forest). It can be concluded that the old forests are the main accumulators of down dead wood biomass and carbon.

Keywords: forestry management, beech forests, biomass, carbon cycling



Section 6

NATURAL RESOURCES MANAGEMENT



Application of Ontologies and Semantic Web for Facilitation of Ecosystem Services Assessment

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The application of Ontologies in the description of biology has become widely spread. This paper presents an approach for the application of Ontology in the description of complex ecological categories as Ecosystem Services. The authors illustrate the application of two Ontology approaches of engineering – usage of Unified Modelling Language and Semantic Web approach – usage of RDF/OWL schema languages. Both approaches are powerful enough to be used as tools for formal description and information dissemination of complex ecological categories. The conversion of both approaches in general is under development in the Object Management Group (OMG).

Keywords: Ecology knowledge, Ecosystem services, Ontology, OWL, Unified Modelling Language, UML

Basic methodological postulates for determining ecological footprint in Republic of Macedonia

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The demand for natural resources in satisfying the needs of humanity is in constant rise. Determining the biological productivity of the land use categories and the anthropogenic influence on them, poses a challenge because of the essentiality of sustainable management of the natural resources in a given geographically defined area. The goal of this research is to determine the extent of the pressure by the anthropogenic factor on the land use categories in the Republic of Macedonia by implementing the ecological footprint indicator. The methodology for calculating the Ecological Footprint Account – EFA on a national level allows for expressing the quantities of anthropogenic influence on separate land use categories, from which the resources for maintaining the needs of the population are supplied. The implementation of the appropriate steps for rational and effective use of natural resources will account for sustainable use of the land use categories and relieve the anthropogenic pressure on them.

Keywords: land use, bioproductivity, human pressure

Sustainability of Forest Ecosystem Services Based on Tree Eustress Nomenclature and Climatic Patterns

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Holistic approach for the trees eustress identification, as statistically proven periodically lowering in radial stem growth that summarized the final result of complex environmental influence, has developed. The major indicator used is the growth index (ratio between measured and calculated value of tree ring width by approximating functions). The methodology is based on dendrochronology analysis and developed software SP-PAM. The tree stund modeling based on the eustress peculiarities and climate is the credible approach for the expected forest changes prediction. The eustress periods have their quantitative markedness - the stem growth reduction, associated with forest community production and the amount of absorbed carbon dioxide and released oxygen reduction. They have also the appropriate quality markedness - frequency, duration and depth, resulted from environmental changes and trees ecological pedicularities. Quality markedness allows to create the eustress assessment scales and nomenclature, also to seek tree functional types (TFT). Selected thus reactive TFT have a direct or indirect connection with forest areas and climate changes.

The results from analysis of *Abies alba* L., *P. sivestris* L., *Fagus silvatica* L. and *Quercus cerris* L. chronologies are presented. The bulk of data is from international databases and from Bulgaria. The relationships between climatic patterns (sequence of climatic types for three-year periods, e.g. stressful year and two years before).

The proposed approach can make a significant contribution to the monitoring and forecast of changes in forest vegetation (species composition, structure, production and occupied area) and climate. The proposed reactive functional types can support future DGVMs development.

Keywords: dendrochronology, growth index, eustress characteristics, climatic type, tree functional type, SP-PAM application

Macedonian face-to-face ethnobotany

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The article examines the relationship between people and plants between three ethnic groups (Macedonians, Albanians and Turks) in Southwestern Region of Macedonia. A number of methods are used, including face-to-face in depth interview with knowledgeable elder's with empirical knowledge; focus group discussion with farmers; face-to-face interview with Protected Areas managers; semi-structured interview with local healers; face-to-face in depth interview with local sellers of medicinal, and food/edible plants; Documented data was evaluated using Fidelity Level; Informant Census Factor and Relative Frequency of Citations.

The finding shows that from the total of 159 reported plants, 112 have medicinal uses, 76 were reported as food/edible and 12 had symbolic use. The most popular ethno-medicinally reported plant species were used for treating diseases related to respiratory and digestive system. Study reveals how one species *Buglossoides purpureoacerulea*, traditionally used to dissolve kidney stones can become very interesting for future phytochemical and pharmacological studies.

Keywords: Macedonia; Ethnobotany; Medicinal plants

Estimating economic value per carp and non-carp fish stocked at Lakes Prespa using non-market valuation models

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The objective of this study was to estimate the change in angler trips as a response to current season stocking level, in order to calculate the net marginal economic benefit per fish stocked at Lakes Prespa. As an alternative, this study aimed also to estimate and utilize the relationships among catchable carp stocking level, angler catch rate, and visitations, in order to calculate the economic benefit of stocked fish. In this study are used in conjunction three models to derive a marginal stocked fish value: a modified version of the Travel Cost model; the Zero Inflated Poisson stock-catch model, which establishes the relationship between stocking intensity and catch rate, and a catch-trip equation which calculates the relationship between catch rate and annual fishing trips. Finally, using all of the above and data from a 2013 angler survey, the method for deriving a marginal value of stocked fish is presented. Results revealed that for both carp and non-carp anglers, there exist positive relationships between stocking intensity and catch rate, and between catch rate and annual angling trips, and that while the average consumer surplus per angler day for carp anglers exceeded non-carp anglers' by almost a factor of three, the final derived net economic value per stocked fish for non-carp is five times that of carp's. This disparity can be traced back to the average daily catch rate, as well as other summary statistics. On average, non-carp anglers took more trips on a year, spend more days on a trip and fished longer on an angler day. Coupled with higher catch rate, it is intuitive that non-carp anglers would respond to the increased stocking level with greater magnitude than non-carp anglers. This translates into the higher predicted annual trips from increased catch, hence non-carp's higher net benefit per fish stocked.

Keywords: angler trip, angler catch rate, carp and non-carp fish, net marginal economic benefit, stocked fish

Some historical data on restocking open waters of Bosnia and Herzegovina

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With the aim of determining the origins, course and further process of restocking the open waters of Bosnia and Herzegovina, there has been access to the articles written on this issue, contained in the paper "Fisherman's Journal". This newsletter was published by National Fishermen Association of Bosnia and Herzegovina, and newsletter archive copies were stored at the public library "Dervis Susic" in Tuzla. By reviewing the above mentioned articles it was established the existence of written information about the beginnings of restocking our waters dating back to 1913. Restocking was conducted discontinuously, insufficiently planned and often without adequate checks of the entered fish materials. In this publication there are data on some restocking of the open waters conducted during 1925, 1947, 1961, 1964, 1965, 1967, 1969, 1971, 1972, 1973, 1976, 1977 and 1983. The most extensive restocking took place during 1965 when released 2,876,489 pieces of different kinds of fish in open water, and the smallest one restocking took place during 1925, when entered 29000 pieces of fish. Discontinuity of restocking was dependent on a variety of factors and was often dependent on the political situation of the Balkan. The waters of Bosnia and Herzegovina during the mentioned period were stocked by salmon kind of fish (trout, salmon, and Arctic char, grayling), cyprinid kind (carp, tench, crucian carp), but also with other kinds of fish from other families such as pike and catfish. Most waters were stocked with trout. Based on collected data, it can be concluded that the waters of Bosnia and Herzegovina were stocked on with indigenous as well as with introduced fish species.

Keywords: restocking, open water, fish, water

Mapping and social valuation of ecosystem services in Bregalnica watershed, Eastern Macedonia

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The research encompasses the concept of ecosystem services intended to reflect the relationship between human population within Bregalnica watershed with nature and the benefits people have from the ecosystems.

By using the method of mapping and non-monetary (social) valuation of ecosystem services we performed an assessment of ecosystem services in the Bregalnica Watershed. Initial analysis for determining the capacity limits of the investigated area are made by using GIS tool. We used data from CORINE Land Cover as a starting point for mapping the capacity to provide ecosystem services. Non-monetary valuation of ecosystems and ecosystem services was done by using questionnaire, which

covered 257 respondents from all populated places of the pilot area. Hence, a selective list of ecosystem services was used which includes locally relevant ecosystems and ecosystem services.

Broadleaf and mixed forests have a very high potential to support biodiversity as confirmed by the presence of certain species. The highest values of the ecosystem services demand can be found in urban, industrial, commercial and plain areas with various crops. Wild and natural land cover types as well as unpopulated high-mountain areas are characterized by an enormous capacity to supply ecosystem services exceeding the demand.

The survey has shown that forest ecosystem is the most valuable due to providing economic benefit, clean air or suppliers of firewood. Wood, medicinal and aromatic plants and water are the most used provisioning ecosystem services.

Gained results can serve as a good basis for a better understanding of the socio-economic situation in the region which will contribute to comprehensive preparation of future development plans. Used concept of ecosystem services follows the world trends which bring us to fulfilment of various targets or actions set by international conventions and strategies in order to enhance the benefits provided from the nature to the people.

Keywords: ecosystem services, valuation, Begalnica watershed

Important Plant Areas conservation in Macedonia in the hands of local communities

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Ever since the 42 Important Plant Areas (IPAs) were identified in Macedonia in 2007, nothing has been done in terms of their conservation enforced by the national legislation. They were only approved as such and mentioned in the national law for nature protection. Thanks to the initiatives raised by foreign organizations, four IPAs related projects have been applied in Macedonia in the period 2007-2016 realized by national and international NGOs.

Especially important is the project known as “A Natural Network for People and Places”, conducted in three phases which covered eight IPAs from Macedonia. These IPAs were selected and networked with a dual objective in mind: to contribute to the protection of plants and raise awareness among the people about the value and importance of nature in general. During this project a network was formed of local NGOs, stakeholders, informal groups and volunteers that actively took part.

In each IPA, the activities were conducted by local coordinators, responsible for organizing around 30 events that included activities, such as, educational camps, setting information panels along mountain trails, photographing wild plants and development of herbaria, establishing botanical trails, monitoring. Some 1700 people were part of the Natural Networks project. Mountaineers, teachers, students, youth groups, representatives of the municipalities - all voluntarily participated in the events.

The transfer of experience and knowledge enhancement on the international laws for nature protection is especially important and beneficial to the local communities that potentially can become so called local conservation groups (LCGs). These LCGs are precisely those target groups that should be familiar with the nature protection laws. Thus, they become directly aware of the importance of

the area/s they act in and at the same time they acquire the capacity to conserve and protect natural resources.

Keywords: Important Plant Areas, Natural Network, wild plants, volunteers

Assessment and mapping of sparsely vegetated land ecosystems condition and their services on the black sea coast, Bulgaria

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Ecosystems with sparse vegetation are natural areas where vegetation is completely absent or is very sparse. This type of ecosystems is well represented along the Bulgarian Black Sea coast and includes coastal dunes and sandy shores, coastal shingle and coastal rock cliffs. Although these ecosystems occupy a relatively small area of the country, they are essential for the conservation of a number of rare and endangered representatives of the flora and fauna of Bulgaria due to the very specific environmental conditions. On the other hand, these ecosystems provide a number of important services to people associated with tourism and recreation, protection of historical and cultural heritage, picturesque landscapes, symbolic and sacred monuments, etc. This study provides information and maps on the status of ecosystems with sparse vegetation along the Bulgarian Black Sea coast, on factors affecting this state, on the services they provide, and the manner and scale of their use.

This work was supported by the Financial Mechanism of the European Economic Area 2009-2014, Contract № Д-33-88/28.08.2015 “Mapping and assessment of sparsely vegetated land ecosystem services in Bulgaria (SPA-EcoServices)”.

Keywords: Bulgaria, ecosystem state, ecosystem services, mapping

Increasing the public awareness for nature protection Survey for determination of awareness of the population for proclaiming National Park Shar Mt.

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In 2015 Macedonian Ecological Society conducted a representative face to face survey for determination of awareness of the population for proclaiming National Park Shar Mt. in the villages

that are within the massive as well as Tetovo and Gostivar urban areas. The scope of the research was to determine the awareness status among the inhabitants that are in this region for proclaiming this massive as National park. This survey is a part of initiative for proclaiming Shar Mt. as National park that is conducted in the past 19 years by the Macedonian Ecological society. The positive effect of the activities through 8 initiatives, 4 programs with international organizations and active cooperation with local NGO's during this period are showing high awareness of the population that are supporting the initiative for proclaiming National park for this massive. Up 82% of the population that inhabits the region of Shar Mt. confirmed the support with 92% urban respondents and 79% of rural respondents. Furthermore 79% of the respondents are having perception that the nature of Shar Mt. is unprotected and 97% consider that nature in general should be protected. The results are showing that there is high general acceptance from the population in Shar Mt. region for the initiative for National park Shar Mt.

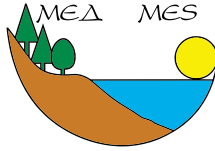
Keywords: survey, protection, park, Shara Mt.

The nature conservation programme in Macedonia

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The Nature Conservation Programme in Macedonia (NCP) is a project of the Swiss Agency for Development and Cooperation (SDC), coordinated by Helvetas Swiss Intercooperation and Farmahem. The Programme intended to support the nature conservation activities and local development of the Bregalnica Region. The main objective of the NCP was to assist the country in conservation of its outstanding biodiversity and natural ecosystems through promotion of their sustainable management and use. The Programme contributed to nature conservation and helped the Republic of Macedonia in facing with NATURA 2000 requirements, one of the pre-conditions for EU accession. This Programme was an umbrella programme that integrated all activities in the Bregalnica Region and practiced holistic approach to the implementation of the interventions for nature conservation and sustainable development.



Section 7

**ENVIRONMENT, POLLUTION AND
GLOBAL CHANGES**



Soil pollution as a consequence of mining and metallurgical activities (study cases from former Yugoslavia)

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The main purpose of this paper is to present geochemical investigations in former Yugoslavia. Intensively, in the last decade Geological Survey of Slovenia (GeoZS) spread the geochemical researches to Bosnia and Herzegovina, Macedonia, Croatia, Kosovo and Serbia through many bilateral and European projects where the mines, smelters and ironworks were the main target of investigation.

This study represents a summary of environmental geochemical researches in the countries formed by breakup of Yugoslavia. Basically, the major goals of GeoZS are strengthening an international scientific cooperation network and partnership with other former Yugoslav countries, improvement of material research standards, exploiting the research and technological demonstration results as well promoting the GeoZS to regional centers of excellence. Our researches are mainly focused on mining and metallurgical processes which are obviously the biggest destructors of environment: B&H (Ironworks Zenica; Ironworks Vareš and Fe mines - Smreka, Droškovac, Brezik, Pb-Zn-Ba mine Veovača); Croatia (the Drava valley; Experimental geochemical map of Slovenia and Croatia); Kosovo (Pb- Zn mine Trepča and Pb smelter Zvečan-Kosovska Mitrovica); Macedonia (Cu mine Bučim, Pb-Zn mines SASA and Toranica, As-Sb-Tl mine Alšar), Pb smelter Veles; FeNi – Kavadarci; thermoelectric power plants (Kičevo and Bitola), alluvial deposits of the Vardar River, Skopje); Serbia (Mine and flotation Bor).

Special attention will be paid to the further development of advanced methods of data processing and the use of methods of linear and nonlinear mathematical modelling: multivariate statistical methods, multiple polynomial regression, and artificial intelligence - Multilayer perceptron. Particularly promising are preliminary results of applications the artificial intelligence methods and different architectures of multilayer perceptron for prediction of spatial changes in contents of chemical elements depending on various spatial and climatic factors that affect these changes.

Keywords: ore processing, contamination; heavy metals, former Yugoslavia

Bioavailability and bioaccumulation characterization of essential and heavy metals contents in various plant food from polluted and referent areas

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Bioavailability of metals occurring in soil is the basic source of its accumulation in plants. The impact of soil pollution (due to urban and mining areas) on the food chain presents a challenge for

many investigations. Bioavailability of metals in a potentially polluted soil and their possible transfer and bioaccumulation in several vegetable species and herbs was examined. Microwave digestion was applied for total digestion of the plant tissues, while on the soil samples open wet digestion with a mixture of acids was applied. Three extraction methods were implemented for determination of bioavailable metals in the soil. Atomic emission spectrometry with inductively coupled plasma was used for determination of the total contents of 21 elements. Significant enrichments in agricultural soil for As, Pb and Zn (in urban area), Cd, Cu and Ni (in a copper mine area), compared with the respective values from European standards were detected. On the basis of three different extraction methods, higher availability was assumed for both lithogenic and anthropogenic elements. Translocation factors higher than 1 were obtained for As, Cd, Cu, Ni, Pb and Zn. Higher root to shoot translocation of these metals indicated that plants species have vital characteristics to be used for phytoextraction of these metals. The obtained data also suggested that *Spinacia oleracea* and *Rumex acetosa* were singled out to have a phytostabilization potential for Cd, Cu, Ni and Pb, while *U. dioica* only for Cu. *R. acetosa* has a potential for phytoextraction of Cd in urban and copper polluted areas.

Keywords: heavy metals, bioacculation, pollution, plant food, contamination risk, Macedonia

Enforcement and compliance of environmental legislation in the Republic of Macedonia

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The Republic of Macedonia's accession to EU is a strategic goal supported by the majority of the citizens living in the country. One of the obligations that the Republic of Macedonia should meet is transposing the EU legislation into the national legislation. In that sense, the area regarding environmental protection has one of the leading roles and it is actually before the rest of the areas. Part of the legislation in this area had been transposed up to year 2005 and it has been being a subject to implementation so far. Implementation and enforcement of legislation have a special place in EU legislation on environmental protection. In the case with the Republic of Macedonia, all this has been covered with the Law on Environment, Law on Nature Protection, Law on Waters and Law on Inspection Supervision. This paper tries to give a detailed analysis of the Macedonian case, shedding a light on the period from 1996 to 2016. In doing so, what is taken into consideration are the inspections carried out on the behalf of the State Environment Inspectorate. The paper also sheds a light on the all aspects regarding the implementation of these laws in practice. The results from this research lead to the conclusion that the implementation, enforcement and compliance have an upward trend of movement, but there is still a room for further improvement.

Keywords: Inspection supervision, European Union, State Environmental Inspectorate, Planning and Reporting, IRAM, BPMS.

Lead content in 15 species of wild fungi in the area of the thermal power plant Oslomej (Kičevo valley, Macedonia)

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The aim of this paper is to analyze the content of lead (Pb) in fruiting bodies of 15 species of wild fungi. Fungi and soil samples were collected on 12 localities at three distances from the Thermal Power Plant Oslomej located in the Kičevo valley in the west part of the Republic of Macedonia. Fungi were collected in spring and autumn during three years period (2012-2014). The average content values of Pb were higher than the European standard of 2.00 mg·kg⁻¹ in all of the analyzed species. The maximal values in some samples of edible species were up to five times higher (*Marasmius oreades* - 11.93, *Russula cyanoxantha* - 9.78, *Armillariella mellea* - 9.52, *Amanita rubescens* - 9.35, *Cantharellus cibarius* - 9.28 mg·kg⁻¹, *Boletus aestivalis* - 8.86, etc.).

In general, statistical analyzes showed no direct correlation between Pb content in the fruiting bodies and total/extractable Pb in the soil samples. The discriminant analyzes proved that the Thermal Power Plant Oslomej has impact on the heavy metals content although direct and significant correlation between the distance from the power plant and Pb content in fruiting bodies was observed in only few species. Nevertheless, the Pb content in the wild fungi in Kičevo valley is very high and the use of the edible species should be controlled and monitored.

Keywords: Fungi, lead, Thermal Power Plant Oslomej, Kičevo, Republic of Macedonia.

The impact of thermal power plant Oslomej (Kičevo valley, Macedonia) on heavy metals content (Ni, Cu, Zn, Fe, Mn, Pb, Cd) in fruiting bodies of 15 species of wild fungi

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Many mushrooms species from the Kičevo valley (Republic of Macedonia) are consumed by the native human population regardless of the heavy metals content and uptake in the human body. The aim of this study was to evaluate the impact of the Thermal Power Plant Oslomej on the extent of heavy metals accumulation in the fruiting bodies of selected fungi.

The material of fruiting bodies of 15 species of wild fungi and soil samples were collected on 12

localities at three distances (0.5-0.75, 2.5-3.5 and 6-8 km, respectively) from the Thermal Power Plant Oslomej in the period from April 2012 to May 2014. We analyzed the content of seven heavy metals (Ni, Cu, Zn, Fe, Mn, Cd and Pb) by atomic adsorption spectroscopy in the fruiting bodies that were not rinsed before drying.

Significant correlation between the distance and content of certain heavy metals was found in 10 fungi species, especially in the cases of Ni and Cu and less in the cases of Cd, Pb and Fe. Such correlations between heavy metals content in soils (total and extractable) and fruiting bodies were found mostly in the cases of Mn, Zn, Fe, Cd and Cu. Also, the Canonical Discriminant Analysis showed possible impact of Thermal Power Plant Oslomej on heavy metals patterns in *Boletus aestivalis*, *Russula cyanoxantha*, *Cantharellus cibarius* and *Hypholoma fasciculare*. It can be concluded that both the pollution from the Thermal Power Plant Oslomej and soil composition determine the heavy metals content in the fruiting bodies of wild fungi in Kičevo valley.

Keywords: Fungi, heavy metals, pollution, Thermal Power Plant, Republic of Macedonia

The content of heavy metals in the soil by the state road Čačak-Kraljevo

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The origin of heavy metals in soil is geochemical. Their concentration caused by anthropogenic influences has increased with the development of industry and agriculture. Soil pollution by heavy metals is a significant environmental problem worldwide. Their toxicity has high negative impact on ecology, nutrition and environment.

Physico-chemical soil analysis is performed in order to determine the presence of contamination caused by agrochemicals from farmland and frequent traffic on the nearby road. Sampling was carried out in April 2016 at three locations (L1, L2 and L3) near state road Čačak – Kraljevo, at the waterfront of tributaries of Zapadna Morava River, at a depth of 30 cm. The analysis of the tested parameters is carried out in accordance with current legislation. Organic matter content, pH and clay content, as well as harmful and hazardous substances (Pb, Cd, Zn, Cu, Ni, Cr, Hg, As) was determined. The contents of Cu, Cr, Ar (L2) and Cr, Hg (L3) were crossing the limits given in the Regulation. The content of Ni (72.1) at the site L1 was higher than the limit value, and L2 (96) and L3 (102) has been over remediation values. Limit and remediation values for L1 was higher (the result of the higher content of clay (14%) and organic matter 6.42%), with respect to L2 (0.31%; 2.78%), and L3 (0.05% and 4%), respectively. Only the content of Ni was higher than limit values of MDK (50 mg kg⁻¹). Based on previous research in this region it can be concluded that the increased content of Ni in the soil of geochemical origin.

Investigated farmland near busy roads is not threatened by Pb. Soil samples did not show acid reaction (obtained value: pH 8.03; 8.52; 8.31), so mobility of heavy metals and the risk of contamination of groundwater not expected.

Keywords: Pollution, hazardous substances, remediation values, MDK, environment

The flora and vegetation of the area around street trees (“Baumscheiben”) from the city of Skopje compared to other five sites

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This study was carried out to determine the flora and vegetation of the area around street trees (“Baumscheiben”), habitat type that is typical urban and which is represented in nearly all cities of the world. The spontaneous flora of this habitat was recorded in six towns (Skopje, Belgrade, Podgorica, Sarajevo, Zagreb and Ljubljana) situated in six countries from the Balkan Peninsula.

The aim of this study is to display characteristics of Baumscheiben vegetation from the city of Skopje compared to the investigated Baumscheiben vegetation of the other five cities.

The flora in more than 50 Baumscheiben relevés was recorded in each of the mentioned cities during May 2016. Thereby, were chosen Baumscheiben that showed no sign of horticultural treatment, covered by at least 30% of spontaneous vegetation consisting of at least five species. Mean indicator values and the mean proportion of each life form were calculated. We used non-multidimensional scaling (NMDS) to test for floristic differences between the Baumscheiben of the six Balkan cities. The species which are dominant and have a high degree of constancy were determined by calculating the constancy of each species to each group (cluster) of relevés of different town, using phi coefficient in the program Juice.

In the 54 investigated relevés from the city of Skopje were found 59 herbaceous species around 11 species of trees, out of which 19 Baumscheiben relevés are around *Tilia tomentosa*. The comparison of the life form show that the Baumscheiben vegetation is therophytic-hemicriptophytic. The most important families are the *Poaceae*, *Asteraceae* and *Brassicaceae*, all more frequently represented. The most dominant species that characterize Baumscheiben vegetation from the city of Skopje are: *Capsella bursa-pastoris*, *Hordeum murinum*, *Stellaria media*, *Poa annua* and *Polygonum aviculare*.

Because the investigated plots are not bound to wet soils, plant species are adapted to high light conditions and to temperatures higher than that of the surrounding area of the city. Also it can be concluded regardless of geographic location there is low variation between the relevés from the different countries it is very likely that homogenization is correlated with increasing urbanization.

Keywords: urban flora, Baumscheiben, Skopje

Spatial Distribution of Heavy Metals in Soil Samples in Mavrovo Region

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The soil is very sensitive medium, an important natural, social and economic resource. The soil can be polluted by various ways. As a biggest and most important factor is human factor. Depending on the nature of the pollutant, soil pollution can be divided into two categories: organic and inorganic pollution. The pollution with heavy metals falls into category of inorganic pollution.

The results from the conducted monitoring of soil in Mavrovo region, Republic of Macedonia, are presented. The content of 19 elements was examined such as Ag, Al, Ba, Ca, Cr, Cu, B, Fe, K, Li, Mg, Mn, Na, Ni, P, Pb, Sr, V and Zn. In total 44 samples were collected from 22 measuring places at depth of 0-5 cm of top soil and at depth of 20-30 cm as bottom soil. Collected samples subsequently were prepared in laboratory with a series of processes (drying, sieving, digestion) to make the samples as solution and to be used in an appropriate devices. The determination of the content of elements was performed by atomic emission spectroscopy with inductively coupled plasma (ICP-AES). After the obtaining of the results, statistic analysis was made for processing the data and appropriate spatial distribution maps for each element were prepared. To determine the origin of analyzed elements, factor analysis was applied. Three factors were obtained (F1, F2 and F3). It was concluded that all the presented elements in that region were with lithogenic origin.

Keywords: soil, heavy metals, Mavrovo, factor analysis

Phytoremediation of cadmium polluted soils using soybean varieties

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Industrialization and extraction of natural resources have resulted in large scale environmental contamination and pollution. Soil pollution with cadmium is due to strengthened industrial development, especially in the areas of drilling, exploitation and processing of mineral raw materials. On the territory of the Republic of Macedonia there are several areas with significant higher content of cadmium in the soil, including the vicinity of the mine lead and zinc "Zletovo" near the town of Probištip. Phytoremediation is one of the most convenient techniques for remediation of heavy metals from contaminated soils. The main purpose of the present study was to determine the effectiveness of soybean varieties for phytoremediation of agricultural soils with higher content of cadmium. For that purpose, three soybean varieties with long vegetation were used: *Balkan*, *Ilindenka* and *Pavlikeni* in association with rhyzobacterium *Bradyrhizobium japonicum*. The total and available content of cadmium were

determined in separate parts of the plant (root, stem, leaf, seed and pod). Physicochemical analyses were conducted for determination soil properties. Bioaccumulation factor (BAF), bioconcentrating factor (BCF) and translocation factor (TF) were used to examine the soybean potentiality for cadmium remediation.

Keywords: cadmium, soybean, phytoremediation, soil pollution.

Protection of population from pollution by lead and other heavy metals in Mitrovica

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Pollution is a global problem which knows no borders, risking three basic elements from which begins life context itself: air, water and land. Kathy paper presented during pollution in Mitrovica, which is known as industrial country, and the industry as Frontline cause pollution poses a very big problem and concern for the local residents of this area. Frontline causes pollution in regions of heavy metals Pb, Zn, Cu, Fe, Cd which negatively affect human health.

Keywords: pollution, heavy metals, Pb, Zn, Cu, Fe, Cd

The disposal of waste to landfill during 4 years in Mirash and impact on the environment

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Landfill of Prishtina presents a series of operational and environmental problems that inhibit the activity of landfill consistently and effectively. Urgent problem requiring immediate solution is flooding the landfill, but certainly there are other problems. The situation in the landfill is serious in all aspects. From the environmental point of view there is a serious risk of pollution of all space including underground waters (IFC2010)

Increasing the number of clients automatically increases the quantity of waste deposited. The use of packaging products and processed them and less of organic products in particular during the season of winter increases the amount of waste and their processing is more difficult ,higher cost and increases the risk of environmental pollution.

Results show that from 2012 to 2015 is to increase the numbers of consumers to 10,049 or 33.81% compared to this we also increase the amount of waste disposed from 2012-2015 to 18,873.00 (T) or 32.48%

This research aims to analyze the change of the amount of waste from year to year based on the increasing number of customers and more negative impact and problems that may arise in the future environment. The data are taken from the Regional Waste Company “Cleaning” JSC - Prishtina on the number of clients and amount of waste disposed through data entry every day during waste disposal in landfill Mirash which is through the measuring scale located at the entrance to the landfill. Physico-chemical analyzes and bacteriological parameters show that most are above the threshold with what tells us about the dangers of environmental pollution and the increasing risk apprenticeship

Keywords: Landfill, Kosovo

The distribution and accumulation characteristics of heavy metals in Macedonian tobacco - comparison with corresponding soil

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The distribution and accumulation behavior of heavy metals is strictly specific, both in terms of plant species, and in terms of the specific element. The main aim of the presented study was to assess the heavy metal uptake and distribution in the different vegetative organs of the tobacco as the most important agricultural plant grown in Macedonia. The study was conducted on three well known tobacco regions in Macedonia (Pelagonia, Southeastern Region and Vardar Valley). Heavy metal composition (Cd, Cr, Cu, Mn, Ni, Pb and Zn) of the oriental tobacco samples and corresponding soil extracts were analyzed using multivariate statistical technique such as principal component analyses, factor and cluster analyses. The following samples were analyzed: tobacco leaves (three harvesting zones), roots, stems, blossoms, seeds and corresponding soil. It was found that the elements distribution is quite heterogeneous in different parts of tobacco plants. The results generally show the highest content of analyzed elements in leaves (Cd-0.41 mg/kg, Cr-2.7 mg/kg, Cu-12 mg/kg, Mn-68 mg/kg, Ni-2.5 mg/kg, Pb-1.0 mg/kg, and Zn – 22mg/kg). Only Cu (15 mg/kg) and Zn (78 mg/kg) had higher content in the tobacco seeds. Availability of studied elements calculated by DTPA extraction and total content of each element in corresponding soil samples is in the following order: Cu>Cd>Mn>Pb>Ni>Zn. The highest Biological Accumulation Factor (BAF) is obtained for Cu, Cd and Zn, showing that oriental tobacco accumulates greater amount of these elements. Analyses of the Biological Transfer Factor (BTF) show that the biggest transfer in the aerial organs of oriental tobacco have Cd and Zn. Furthermore in the sequence follows: Mn>Cr>Cu>Pb>Ni. Despite intensive tobacco production, only few soil plots

had high Ni content, all other analyzed elements from soil and plant samples were at levels which are typical of agricultural and low anthropogenic pressure areas.

Keywords: oriental tobacco, pollution, agriculture areas

Heavy metals content in soil and fungi of Skopje urban area

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The main aim of this investigation was to determine the Pb and Cd contents in some lignicolous and terricolous fungi from the urban part of Skopje city (the yard of the Institute of Agriculture). The content of Pb and Cd was determined in the wood of *Acer negundo* and *Ailanthus altissima*, the substrate of the lignicolous fungi (*Flammulina velutipes*, *Auricularia auricula-judae* and *Agrocybe aegerita*). Soil samples were collected from soil pits at depths of 0-30 and 30-60 cm.

The digestion of the soil samples was performed in concentrated HCl and HNO₃ in a ratio 3:1; digestion of fungi and wood was performed with concentrated HNO₃ and H₂O₂ in a Heating Digester DK 20. The quantity of heavy metals was determined by use of atomic absorption spectrometry on Agilent 55 and Agilent graphite furnace 240Z.

The results of the analyses showed that higher Pb and Cd contents are present in the soil samples under *Ailanthus altissima* compared to the ones under *Acer negundo*. In contrary, the contents in wood were higher in *Acer negundo* compared to *Ailanthus altissima*.

The contents of Pb and Cd in the terricolous fungus *Lepista nuda* were higher than in the analyzed lignicolous fungi. The content of Pb was almost as twice as higher than the maximal levels for food contaminants in all of the analyzed fungi. The content of Cd was lower than the maximal levels with the exception of *Lepista nuda*. The lowest Pb and Cd contents were determined in *Flammulina velutipes*.

Keywords: heavy metals, soil, lignicolous fungi, terricolous fungi, wood

The comparison of Mn, Fe, Zn and Pb contents in substrate and fruiting body of Amanita taxa from Samanlı Mountain (Marmara Region-Turkey)

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Mn, Fe, Zn and Pb contents in substrate and fruiting body of *Amanita* taxa from Samanlı Mountain (Marmara Region-Turkey) were compared. Compared *Amanita* taxa are *A. caesarea*, *A. excelsa*, *A.*

franchetii, *A. gemmata*, *A. mairei*, *A. muscaria*, *A. pantherina*, *A. phalloides*, *A. rubescens*, *A. vaginata* and *A. verna*. Mn, Fe, Zn and Pb analysed by ICP-AES equipment. Data were compared statistically by Correlation Test. Mn and Fe content in the likely negative correlation was observed in Zn and Pb contents likely positive correlation. The correlation coefficient (r) -0.5094 for Mn, Zn; 0.02186, -0.3457 to Fe was calculated as 0.20303 for Pb.

Keywords: substrat, fruiting body, Amanita taxa, Mn, Fe, Zn, Pb

DNA damage and histological changes in gills and liver of cyprinid fish as biomarkers of the Sava River pollution

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Fish species living in freshwater ecosystems are exposed to a various range of xenobiotics. Two of the most important pathways of xenobiotics intake by fish are either via the digestive or respiratory routes or by absorption of waterborne chemicals through the gills. As a biomarker of exposure to genotoxic agents, the single cell gel electrophoresis (SCGE), the comet assay, is widely used for detecting DNA damage in different tissues of aquatic organisms. As biomarkers of effect, histopathological analyses are considered efficient and sensitive for monitoring the fish health and pollution of aquatic ecosystems. Examination of multiple biomarkers in exposed organisms may give important information on organisms' exposure to xenobiotics and related stress response.

The main objective of this study was to evaluate the level of the Sava River pollution in the Belgrade region by measuring genotoxic and histopathological biomarker responses in liver and gills of cyprinid fish. Specimens of *Abramis brama*, *Blicca bjoerkna* and *Ballerus sapa*, were collected in winter, spring, summer and autumn of 2014 from the site Duboko. This site is exposed to untreated urban wastewaters, agricultural and industrial activity. DNA damage in liver and gills is expressed by Tail Intensity (TI), while histological changes were expressed as organ index which represents the extent of organ injuries. The highest level of DNA damage in liver and gills was detected in summer. The lowest level of DNA damage in liver was observed during spring and in gills during autumn. HP index of liver (IL) and gills (IG) showed no significant difference between seasons, but IL showed gradual increase from winter to autumn and IG showed a gradual increase from winter to summer. In conclusion, summer is marked as a season in which fish are under higher pressure of pollution according to both DNA damage and histopathological changes in tissues.

Keywords: comet assay, histopathology, Sava River, biomarkers, freshwater fish

The effect of metal exposure on melanomacrophage aggregates and their pigment content in the spleen of the Vardar chub (*Squalius vardarensis*)

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Macrophage aggregates (MACs) are pigment-containing cells accumulations that are presented in fish spleen, kidney and liver. Their number, size and pigment content may change depending on environmental degradation and therefore can be used as bioindicators of water quality. There are some indications that metals have inhibitory effect on fish immune system, but little is known of the effect of complex mixture of pollutants on MACs pool in fish spleen. In this study quantitative influence of heavy metals pollution on the MACs amount in the spleen tissue will be evaluated for the first time.

For this study female (n=26) and male (n=30) Vardar chub (*Squalius vardarensis*) were collected at three rivers: Bregalnica, Zletovska and Kriva Reka, with difference in pollution impact; organic, heavy metals, and mixture of both, respectively. Using stereological technique, relative (Vv) and total volume (V) of the MACs were estimated as well as the amount of the pigments within MACs.

The results showed that when V and Vv of MACs and the contributing pigments in the spleen were estimated, male fish appeared more impacted by pollution compared to female and also showed significant decrease of V of MACs in Zletovska compared to Bregalnica. Moreover, statistically lower value of Vv and V of MACs containing melanin/lipofuscin pigments were recorded in Zletovska when compared to the two other sites. In comparison, Bregalnica, which has mainly organic pollution impact, had highest values of V of MACs compared with other two rivers. Male fish from Kriva Reka showed low V of MACs, with the lowest amount of MACs which contains haemosiderin/lipofuscin pigments. Female fish showed lowest Vv of MACs in Kriva Reka which can be attributed to the significantly lower Vv for the lipofuscin containing MACs in the same locality.

In conclusion, pigment content, as well as the amount of MACs in the chub can change concerning the type of pollutants. Metals reduce the appearance of the MACs pool, mostly through reduction of lipofuscin and melanin/lipofuscin pigments, and probably induce immunosuppression in these cells.

Keywords: pollution, bioindicator, stereology, immunotoxicity, melanin, lipofuscin, haemosiderin

Evaluation of the histopathological alterations in the gills of the Prespa barbell (*Barbus prespensis*) inhabiting a polluted area of Lake Prespa

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The investigation will focus on gill lesions observed in Prespa barbel individuals caught from a north littoral site of Lake Prespa situated between Sirhan and Asamati in the area of tributary Golema Reka entrance, which is under agricultural, industrial and urban pollution pressure. The gill's tissue was analyzed by light microscope on standard paraffin embedded and resin embedded specimens. The quantification of the histopathological condition of the gills was made according to standard protocol for fish health that inhabits fresh water. The number of mucous and chloride cells in gill tissue were determined by histometric analysis. In parallel with this a scoring of present infections was made. Analyses of the gill tissue showed a series of circulatory, regressive, progressive and to a smaller extent inflammatory changes from which the most prominent were: telangiectasis, necrosis of respiratory epithelium accompanied by collapsed secondary lamellae, necrosis and proliferation of interlamellar and lamellar respiratory epithelium, and parasite infection. Quantitative and statistical analyses of the results indicated that the reiterated gills lesions have occurred by double influence, toxic effects of polluted aquatic environment and opportunistic infections. The output of bivariate correlations analyses suggests that the toxic effects from the pollution are dominant.

Keywords: gill, histopathology, pollution, barbel

The effects of waste water treatment through physical and chemical parameters scanning on littoral part OF THE Ohrid Lake (Albanian part)

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The water quality of Ohrid Lake (Albanian part) is analyzed continuously priory and after the operational phase of the wastewater treatment plant. The wastewater treatment plant of Pogradec city started to operate partially in the year 2009. The major goal of this paper is to evaluate the water quality of the littoral zone of Ohrid Lake (Albanian part), six years after the establishment of this wastewater treatment facility. In order to achieve that, some DBO₅ analyses were carried out in the two sample stations. The first sample station is at the end of the wastewater treatment plant and the second one is in the lakeshore which is around 1 km far from the wastewater treatment plant discharge site. In the same time some bacteriological analyses in five sampling localities of the littoral zone were conducted, too. The comparison of these data taken during 2011, 2013, 2014 and 2015 with those before 2009 and with the international standards shows that in several sites the water quality is improved, while in some

others the pollution level is still to be of concern. The treated wastewater quality is improved but the impact of some other factors that are not directly connected to the wastewater treatment plant operation has to be considered further on. Finally, in order to improve the water quality in the polluted sites some recommendations are presented as well.

Keywords: Ohrid Lake, coliform pollution, wastewater treatment plant, wastewater quality, DBO5.

Assessment of the faecal contamination along the Sava River and identification of pollution sources

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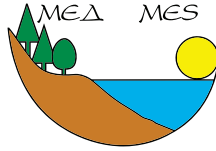
The contamination of water by faecal pollution leads to exposure to pathogens via drinking water production, recreation or irrigation. However, monitoring of microbiological quality of surface waters is quite neglected despite its importance for human health. In the case of Sava River Basin, many of the settlements situated on the river banks discharge high quantities of untreated or improperly treated wastewaters directly into surface waters. Due to usage of water for irrigation, the evaluation of microbiological quality of the Sava River becomes essential for further river management.

Water samples were collected during September 2014 on 17 sites and during September 2015 on 15 sites situated along the Sava River. In 2015, additional samples were collected from 4 wastewater outlets detected onsite. Microbiological analyses comprised monitoring the standard indicators of faecal pollution within the surveys and long term monitoring data (obtained within 5 years of routine monitoring at 4 stations). For detection of total coliforms, *Escherichia coli* and enterococci, Defined Substrate Technology (DST) was used with quantification performed by Colilert Quanti-Tray 2000 system, which provides a Most Probable Number result. Detection of presumptive *Clostridium perfringens* was performed by membrane filtration method according to ISO 14189:2013.

To identify the origin of pollution, microbial source tracking (MST) analyses were employed based on the human-associated BacHum and HF183II, the ruminant-associated BacR and the pig-associated Pig2Bac genetic *Bacteroidetes* faecal markers.

Microbiological indicators showed the existence of hotspots of faecal pollution in the Sava River. MST confirmed that the pollution is human associated. Long term data at selected sites indicated persistent faecal contamination which leads to conclusion that the sites are under the impact of continuous discharge of wastewaters.

Keywords: coliforms, *E. coli*, microbial source tracking, surface water contamination



Section 8

**URBAN ECOLOGY AND HUMAN
HEALTH**



Geographical distribution of disease in the world

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This paper elaborates the geographical aspects of the emergence and spread of diseases in the world. Regional geographical overview of distribution of diseases in the world regarding to natural (terrain, climate, hydrographic and biogeographic) and social factors was made. The paper considers the environmental changes caused by extensive human impacts on the environment.

The aim of the paper is contribution to more open publicity approach regarding the autochthon diseases in certain regions as well as their spreading regarding the technological development and population dynamics in the world.

Keywords: medical geography, the distribution of diseases, the spread of diseases, environmental aspects.

Monitoring of air quality in Drenas in 2015

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The industry development of different economic operators is one of the main causes of pollution, which day-by-day is becoming an alarming issue. Despite operating, the economic operators are non-conforming to different European standards, and they are the main pollutants of essential elements of life such as air, water and soil. Since air pollutants are limitless, the greatest pollutants are; volatile organic compounds (VOC), CO₂, NO_x, CO, sulfur compounds SO₂, PM₁₀, PM_{2,5} etc. In this research we have presented the monitoring of air quality in the region of Drenas where the analyses of air quality are taken from KHMI. The measured parameters throughout the year 2015 are: SO₂, CO, NO₂, and O₃, PM₁₀ and PM 2.5, all these being measured in µg/m³, and always referring to the directive 2008/50 / EC and the Law on Air Protection from pollution (no. 03 / L-160).

Keywords: pollution, pollutants, CO₂, NO_x, CO

The state of ambient air quality in the city of Nis, Serbia

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Numerous epidemiological studies have indicated that air pollution is a significant risk to the environment, quality of life, and health of the population. In that sense, systematic monitoring of ambient air quality is of paramount importance for determining ambient levels of pollutants so that they can be related to adverse effects on man and environment. Monitoring of outdoor air pollution in Nis, Serbia started from 1965 and the great efforts have been made since then to improve the air quality. The objective of this study was to analyze the changes in ambient air quality in the city of Nis since the 1992's. Only three of the major air pollutants, sulphur dioxide (SO₂), soot and dustfall, were considered for the assessment. Ambient air quality was monitored over the last thirty years along with micrometeorological data and the results are discussed. In the most polluted station, the maximum concentration of SO₂ was 163 µg/m³, maximum soot concentration was 77 µg/m³ and dustfall-1163 µg/m³. The results showed that different pollutants displayed two trends. The possible causes of detected air quality trends were also discussed in this paper. In conclusion, air pollutant concentrations in Nis, Serbia are generally worse than ambient air quality standards for USA and EU. Much more long-term studies are needed to assess related public health impacts, and explore mitigation approaches.

Keywords: urban air quality, air pollutants, Sulphur dioxide, soot, dust fall, volatile organic carbons, continuous monitoring

Monitoring of air pollution in the Pirot city (southeastern Serbia) 2002 - 2014

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Air pollution exhibits many negative consequences for living beings. The strongest effects of air pollution are felt in urban areas. This research was done in 2002 for the first time, then 2009 and 2014. Epiphytic lichens are determined and collected from the bark of various species of trees at the level of 1.5- 2 m above the ground, exclusively from the trunks angled no higher than 5°. The aim of the study is to present the changes in air quality over a certain time interval. Using the index of atmospheric purity - IAP values, it has been found that there are different air pollution zones: "struggle" zone and "lichen desert" zone. The "normal zone" has not been observed. Based on research from 2002, 2009 and 2014, comparing the results, it was concluded that the zone "lichen desert" increasingly present as a result of intense air pollution in Pirot. The air quality of the investigated area was also monitored for specific pollutants such as SO₂, NO₂ and soot, by the Institute of Public Health in Pirot. This is the first overview of investigation of air quality on the territory of the city, comparing the results obtained by biomonitoring and physico-chemical monitoring.

Keywords: lichen, air quality, urban area

Macrofungi in urban environment: Case study of Botanical Garden “Jevremovac”, Serbia

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Two-years-monitoring of an outdoor area and the greenhouse of the Botanical Garden “Jevremovac”, resulted in a total of 124 macromycetes species, among which 22 species are firstly recorded for Serbia. The most common species belong to the phylum Basidiomycota (113) and only 11 to phylum Ascomycota (mostly noted in the greenhouse). Saprobs are dominant with 81.5%, 45.2% are lignicole and 36.3% are terricole, parasitic species are presented with 13.7% and mycorrhizal species with 4.8%. *Celtis occidentalis*, *Corylus colurnoides*, *Diospyros virginiana*, *Koelreuteria paniculata*, and *Sophora japonica* as alochthonous plant species were hosts or substrates for development of lignicole fungi which fruiting bodies were noted for the first time at these plants in Serbia. Inedible species are dominant (70 species), 34 species are edible, 5 are conditionally edible, 8 are poisonous and one is hallucinogenic (*Psilocybe cubensis*). Significant number of species belongs to the category of medicinal mushrooms. These species have been used for thousands of years in traditional medicine of Far Eastern nations. Current studies confirm and explain experiential knowledge and discover new species which produce biologically active compounds with antimicrobial, antioxidative, genoprotective and anticancer features. Among species collected in Botanical Garden “Jevremovac” medically significant are: *Armillaria mellea*, *Auricularia auricula-judae*, *Laetiporus sulphureus*, *Pleurotus ostreatus*, *Schizophyllum commune*, *Trametes versicolor*, *Ganoderma applanatum*, *Flammulina velutipes* and *Inonotus hispidus*. Some species, such as *T. versicolor* and *P. ostreatus*, also have the ability to degrade highly toxic phenolic compounds and can be used in soil remediation which is ecologically and economically justified process.

Keywords: Botanical Garden “Jevremovac”, diversity, macrofungi.

The urban environment in the Balkans as a main pathway of the introduction of invasive alien species

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Every year the problems of the urbanization of the environment become more acute. In the process of the forming of the “urbanocenosis” a lot of the indigenous species disappear, and many allochthonous elements invade and spread in the cities. This process causes suspension of highly specialized and / or rare and endemic species, and consequent increase in number of the widespread ones with higher

ecological plasticity. Thus fauna of different cities are usually more similar to each other than the fauna of each city with its fauna of non-urban surroundings. This is especially well seen in areas with high biodiversity as the Balkan Peninsula.

Our study of new and previous records, the pace of distribution and the biological peculiarities of some invasive alien species of Insecta: Heteroptera [*Corythucha arcuata* (Say, 1832), *C. ciliata* (Say, 1832), *Stephanitis pyrioides* (Scott, 1874), *Belonochilus numenius* (Say, 1832), *Oxycarenus* (s.str.) *lavaterae* (Fabricius, 1787)], Diptera [*Megaselia scalaris* (Loew 1866), *Dohrniphoracornuta* (Bigot in de la Sagra, 1856)], Lepidoptera (*Cacyreus marshalli* Butler, 1898) and Mollusca: (*Arion* cf. *vulgaris*; *Arion fasciatus*). Recently established populations of alien species in urban environments may become a reservoir of individuals ensuring their future successful invasion.

Environmental and Biological factors causing the degradation of mural frescoes at the Albanian coastal church of St. Mary of Bishqethem in Lushnja

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Following different approaches to the factors that cause degradation of the mural frescoes in Albanian coastal churches (St. Mary of Bishqethem) it might be confirmed that there is a combination in between environmental and biological factors, along with human negative interventions and lack of maintenance. The most important causes for the mural painting state of degradation were the environmental and climatic attributes like content of oxygen in the air, different air pollutants, light directions and changes in temperature accompanied with changes in humidity completed by the lack of maintenance, the humidity that is active in all its forms as infiltration, ground water and condensation and water leak determined the appearance of salts incorporation, as well as biological growth, these conditions have had a devastating effect.

A survey of biodeterioration phenomena was performed in mural painting at the church dome by swap technique. For each sample, 1g was diluted with 9 ml of sterilized distilled water. Samples were shaken vigorously to form uniform solution of 10-1 concentrations. The analyses of isolated fungi and microbial population in the original compost sample were confirming that after humidity these are the main cause of degradation.

Keywords: biodegradation, Saint Mary church, deterioration, analysis, frescoes, fungi

Wood Inhabiting Fungi and Molds on Cultural Monuments in the Republic of Macedonia

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Research on the fungi of the cultural heritage in the Republic of Macedonia was conducted from 2012 to 2016. The expertise was focused on the biodeterioration caused by wood-inhabiting fungi and moulds. A total of 88 cultural monuments, including 56 monasteries and churches, old houses under protection, mosques and two fortress, were inspected in the areas of the towns of Skopje, Tetovo, Kumanovo, Ohrid, Struga, Strumica, Stip, Kriva Palanka, Prilep, Demir Hisar, Debar, Kichevo, Kratovo, Berovo, the region of the Lake Prespa and the Mariovo area. Most of the monasteries are under state protection. Thirty-five of the inspected monuments had fungal damage. A total of 122 fungi species belonging to 63 genera were identified on the constructive and decorative materials of the monuments. Fungi determination was done in Mycological laboratory, Faculty of Natural Science and Mathematics – Skopje. Wood-inhabiting fungi were found both on the indoor wood (ceiling, stairs, roof inner portions) and external woodwork (gateway, bridge, outer door, roofs, stairs, bell towers). Decay fungi dominated in the roof constructions of the inspected buildings. Most of the wood used in construction belong to deciduous trees (*Fagus*, *Quercus*, *Populus*, *Tilia*, *Acer*, *Juglans*, etc.). The majority of the species are from the phylum Basidiomycota, more specifically from the genera: *Athelia*, *Hyphoderma*, *Hyphodontia*, *Ceriporiopsis*, *Coniophora*, *Gloeophyllum* and *Phanerochaete*. Most dominant species is *Hyphodontia crustosa* (Pers.) J. Erikss. The majority of the identified basidiomycets in Macedonian cultural heritage sites belonged to white-rotters (80%), and the remaining (20%) to brown-rotters. In several cases, mould contamination was noted on wall paintings. Fresh fungal damages found in monasteries and churches are dangerous for wood constructions and frescoes, and they must be eliminated.

Keywords: mycodiversity, cultural heritage, lignicolous fungi.

Severe biological colonization of Monument to Despot Stefan Lazarević (Mladenovac, Serbia)

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Biological growth on historic masonry and monuments is a complex issue, and nowadays very important for conservation science. Bacteria, cyanobacteria, algae, fungi, lichens, mosses and higher plants form a complex ecosystem on monuments' surface and due to their biological activity stone

surfaces can be deteriorated leading to structural and aesthetic impairments. Monument to Despot Stefan Lazarević, located in the village of Crkvine by Mladenovac (Serbia), was protected by Republic of Serbia as the Monument of Culture of Exceptional Importance. Main goal of this research was to describe the community of stone dwellers via visual observation, microscopic and cultivation methods prior to planned conservation treatments of the monument. Visual observation of the monument revealed the high coverage of marble surface with numerous lichen thali of *Physcia* sp. and *Xanthoria* sp., and cushions of *Grimmia pulvinata* moss, which demonstrates high level of biocolonization and deterioration process in progress. Direct microscopy techniques along with cultivations of microorganisms detected the presence of cyanobacteria *Gloeocapsa violacea* and *Nostoc* spp. along with fungi *Alternaria alternata*, *Cladosporium* spp., *Epicoccum nigrum*, *Fusarium* sp., *Gliomastix murorum*, *Phoma glomerata*, and *Rhodotorula* sp. forming dense biofilm coverage on the marble surface. Thick biofilm layer changed the appearance of marble surface causing discolorations in form of black and salmon-pink patinas, probably as a direct result of pigment production by microorganisms. This survey demonstrates the importance of multidisciplinary concept in the research aimed at understanding complex process of stone artifacts deterioration, so adequate conservation measures could be applied.

Keywords: biodeterioration, biological colonization, cyanobacteria, fungi, stone

Presence of toxigenic and pathogenic fungal spores in indoor air of Central Institute for Conservation in Belgrade

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In order to determine total mycobiota, with special attention to toxigenic and pathogenic fungi, indoor air of 20 rooms in Central institute for Conservation in Belgrade was investigated. Volumetric air sampling was carried out during all four seasons along with temperature and relative air humidity measurements. Spore load in the air of individual rooms varied from 20 CFU m⁻³ up to above 26000 CFU m⁻³, with air in majority of the rooms estimated as highly contaminated. Documented CFU m⁻³ of air above 500, presenting a high risk for human health, was documented in 15 rooms in December, 13 in March, 19 in June and in all 20 rooms in September. The highest concentration of fungal spores, documented as a CFU m⁻³ average for all investigated rooms, was documented in June (9596 CFU m⁻³) and lowest in March (1795 CFU m⁻³), correlating with temperature and relative humidity measurements. Various toxigenic and pathogenic fungi were isolated including species of genera *Aspergillus*, *Aureobasidium*, *Chrysonillia*, *Fusarium*, *Penicillium* and *Trichoderma*. Toxigenic and pathogenic species of genera *Aspergillus*, including *A. flavus*, *A. niger*, *A. ochraceus*, *A. versicolor*, *A. restrictus* and *A. calidoustus*, were isolated from the air of investigated rooms, during all four seasons. Allergenic fungi also included *Cladosporium*, *Mucor* and *Rhizopus* species. Although concentrations of pathogenic and toxigenic spores varied, presence of certain fungal spores in indoor air is considered a health hazard, regardless of their concentrations.

Keywords: air sampling, *Aspergillus*, fungal spores, indoor air, mycotoxins, mycoses

Repeated exposure to acute heat stress causes changes in serum activity of AST, ALT and CK in rats

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Exposure to various forms of stress causes cellular damage and increased permeability of the cell membrane, which leads to leakage of cytoplasmic enzymes into the bloodstream. Heat stress (HS) can cause biochemical changes in the activity of aspartate aminotransaminase (AST), alanine aminotransferase (ALT) and creatine kinase (CK) in serum. Since the activity of these enzymes is extremely high in the cytosol of myocardial tissue, the disturbance in their serum activity can reflect the extent of tissue damage due to thermal injury. The aim of this study was to observe the biochemical markers of tissue damage (AST, ALT and CK) in rat's serum during recovery after single or repeated exposure to heat stress ($41\pm 0.5^\circ\text{C}/45\text{ min}$).

For the purpose of the experimental work were used 47 male laboratory Wistar rats, divided in 8 groups. Control group was kept at room temperature. Three groups were exposed to single HS, allowed to recover at room temperature for 24, 48 and 72 hours, respectively. The other groups were exposed to two consecutive HS (24h and 72h after the first HS), recovered at room temperature for additional 24h and 48h, respectively.

Single exposure to HS caused significant ($p < 0.050$) reduction of AST (-26%), ALT (-14%) and CK (-63%) activities, after 24h recovery. The reduced enzymatic activity is maintained through the recovery (48h) and after 72h these changes gradually normalized. Repeated exposure to HS (both 24h and 72h after the first HS) caused increased activity of AST and CK, while ALT activity was decreased. Still, the obtained changes are more evident in animals exposed to second HS, 72h after the first HS. We also found significant coefficients of correlation between estimated parameters.

In conclusion, the results show that both, single or repeated exposure to HS causes significant modification on serum biomarkers indicating tissue damage in heat-exposed rats.

Keywords: Heat stress (single and repeated), AST, ALT, CK, heart, rats.

Ambrosia artemisiifolia L. registered in the Republic of Macedonia

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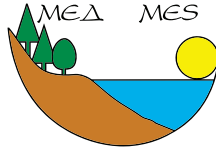
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Ambrosia artemisiifolia is invasive species and strong allergen that is a major threat to the environment as an ecological, agronomic and economic, but also health problem from allergologic aspect. It is naturally distributed in North America, but as an adventive plant incorporated the territory of

South America, Europe, Asia, Africa and Australia. On the territory of Europe this species is widespread in much of the continent and mostly as domestic plant. Detailed field investigations of specific sites to determine its distribution, habitat characteristics and vitality of the discovered populations were conducted. Analysis and monitoring of concentrations of pollen in Skopje (42 ° 01 'N, 21 ° 27' E, 275 m asl) are performed with the volumetric method (Hirst 1952) using a Lanzoni VPPS 2000 seven day sampler in the period of 2003-2011. Chronological studies in the Republic of Macedonia confirmed the presence of *Ambrosia artemisiifolia* in Skopje (settlement Ilinden, side of a highway by hotel Bellevue) in ruderal place together with *Urtica dioica*, *Artemisia vulgaris* and *Chenopodium album*. The number of specimens from *Ambrosia artemisiifolia* rises beside the highway and nearby farmland (planted with peppers, tomatoes and corn) from year to year. The highest concentration of pollen per year was registered in 2004 - 362 pg/m³. Similar annual concentrations were registered in 2006 (357 pg/m³) and 2009 (307 pg/m³). Retrospective analysis and research conducted at the Institute indicate a low prevalence (4.1%), but confirm fierce allergenicity of this kind, among respondents in the Republic of Macedonia (intense skin reactions, ie greater than 4 and specific IgE class greater than 45 IU/ml). In recent years, leading global campaign to find ways to control the spread and suppression of *Ambrosia* in newly settled areas. Our local planning lead to integration into European and worldwide programs to control invasive species and we are include in the project Sustainable management of *Ambrosia artemisiifolia* in Europe (SMARTER)

Keywords: *Ambrosia artemisiifolia*, invasive species, flora, aeropalinoLOGY, pollen, allergen, Macedonia.



Section 9

ECOLOGICAL EDUCATION



Creative Center Karposh – Center of Youth Excellence in the Environmental Sciences and a Tool for Eco-Education

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Creative center Karposh (CCK) is a unique case in Macedonian education system with the mission to provide conditions for interested pupils from the primary schools belonging to Karposh municipality to spend their time out of the classroom in the field of their interest. CCK is composed by ten units that exist in the frame of ten primary schools. The units are organized as a synergy of scientists from appropriate fields, young animators, motivated teachers and interested children. The main principle in the functioning is mobility and possibility for circulating of children according their interest nevertheless which unit exists in their own school. All units are closely related to environment and all of them in their programs have a part acknowledged to eco-education. So, in the Climate changes and energy saving unit, children are discovering phenomena of global climate changes and clean alternatives in energy producing. Design and robotics unit is dealing with eco-design and usage of natural materials. Environmental protection and recycling unit most of its activity is performing on waste management and re-usage of materials. Audio-visual media is serving as an antenna of all CCK activities including those in the field of ecology. Cultural heritage unit is partially focused on the natural history in Macedonia. Units for zoology and for botany & mycology are developing a culture of biodiversity protection among the children. Threw the Food production and control unit the pupils are involved in production of healthy food and its control. Handicrafts unit is working mostly with natural and recycled material and finally Space science unit is interested in global pollution. All those activities are guaranteeing that via CCK mentioned issues will be brought closer to the younger generations and it will provide environmental friendly coexistence between the nature and civilization in this part of the Planet.

Keywords: extracurricular, gifted children, primary schools

Classification of mountains in the Republic of Macedonia in the educational system

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The paper aims to point toward the improper classification (genetic and evolutionary) of mountains in the Republic of Macedonia at all levels of education: primary, secondary and higher education.

Specifically, in geography textbooks in the Republic of Macedonia for primary, secondary and in some for higher education, the mountains in the Republic of Macedonia are divided into „old“ and „young“ (evolutionally), whereas according to the mode of origin (genetically) they are divided into „rocky mass“ и „range“ (chain) mountains.

The classification is inappropriate given that all mountains in the Republic of Macedonia have been created concurrently (with the Alpine orogeny and neotectonic processes), and they all belong to the type of horsts i.e. „rocky mass“ mountains. The aforesaid improper classification has also been incorporated into numerous and diverse ecological studies dealing with mountains, which are a prominent segment of still nature.

Keywords: mountains, Republic of Macedonia, education

Ecological education in Bulgaria – trends and new challenges

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The negative effect of the worldwide society economic development are expressed in food and energy resources reduction of the Earth, disturbed ecological balance, ill people, etc. Predictions of scientists as apocalyptic as they are related to air pollution, the pollution of the soil and the ocean are now a reality and no educated man should doubt their objectivity. This led to a shift in the concept of “unlimited growth” at all cost from the concept of “sustainable development”, “development as coevolution”, etc. The main goal of the material presented is to bring top trends to environmental education in Bulgaria at the beginning of the XXI century, educational policy and research line. To realize the target, teh analysis of the content of 56 articles published from 2011 to 2015 in four Bulgarian educational journals: “Chemistry: Bulgarian Journal of Science Education”, “Strategies for Policy in Science and Education”, “Bulgarian Physics Education” and “Continuing Education” was made. A methodology for content analysis of the selected journals is described using quantitative and qualitative parameters. The results of this study found that most of the published articles were related, their goals are to justify the interdisciplinary nature of the environmental education and training, complex scientific approach implementation to realize the goals of the environmental education, educational practices implementation in united efforts of environmentalists, educators, methodologists and teachers in solving problems related to the protection of the environment, etc.

Keywords: ecological education, journal content analysis, trends in research.

Establishing educational trail in natural environment as a tool for enhancing environmental education

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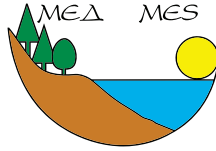
Outdoor education is a priority in understanding the nature and processes that are occurring within. The research presented in this paper was focused in beech forest ecosystem.

The primary objective of this development-action research was to promote the environmental education by monitoring of the impact of indoor and outdoor educational process. Interactive activities and content-related functions, values and benefits of beech ecosystem were developed. An educational trail on Ponikva, Osogovo Mts. was established for this purpose. A survey with questionnaires and interviews was conducted. The study involved 150 students from two primary schools at the age of 10-12 years.

The obtained indicators are realistic and objective concerning the established education, and they confirm the general presumption that the relevant relation and mutual complement of activities and content realized indoors and outdoors give positive results in education. The trail is accepted as a good model that can contribute to the understanding of forest ecosystems and enrich the curriculum in practice.

Education in nature had a positive effect on raising the level of the knowledge and development of environmental awareness among students (indicated by the results of the analysis by the questionnaires) and facilitated their cognitive development (expressed through the creation of literary and artistic works and research interest in nature). Through the character of woodpecker - Vane, the presented content was easily accepted. During the implementation of the activities we proved that the development of such content can improve teaching in nature.

Keywords: environmental education, educational path, outdoor education



Section 10

BIOLOGY STUDENTS' SYMPOSIUM



Ecological status assessment based on aquatic macroinvertebrates - the Belicka River (R. Macedonia) case study

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The aim of the current study was a rapid assessment of the ecological status of river Belica, based on the macroinvertebrate fauna. Belicka belongs to Treska river basin and presents mid-altitude, calcareous river in the 6th ecoregion (Hellenic western Balkan). The material was collected from five sampling sites on the Belicka River during a sampling campaigns in April and August 2016. Standard methodology for collection of bottom fauna (EN ISO 10870: 2012) was followed. The following metrics were used for status assessment: EPT (Ephemeroptera, Plecoptera, Trichoptera) taxa richness, Biological Monitoring Working Party Score (BMWP) and Average Score Per Taxon (ASPT). Survey on macroinvertebrates from Belicka River confirms that larvae of aquatic insects and amphipod crustaceans were principal components of the benthic community. It should be noted that the river is inhabited with important macroinvertebrate species for protection, such as Macedonian endemic gastropod *Radomaniola curta kicavica* as well as the rare caddisfly *Thremma anomalum*. This species were never recorded in 7th ecoregion in the country, which should be taken into consideration in defining reference community for mid-altitude, calcareous river in the 6th ecoregion. In the upper parts of the river the status varied from high to good. Deteriorated ecological condition (moderate toward poor) was observed downstream of the village Belica and fish ponds indicated that the river in this stretch is under significant anthropogenic pressure and contamination with organic compounds. Further work on the ecological status assessment hinges on the development of a regular seasonal sampling campaign, selection of appropriate metrics and development of type specific reference conditions.

Keywords: macroinvertebrates, Belicka River, R. Macedonia, ecological status

Contribution to the knowledge of Coleoptera fauna of Skopje valley and Vodno Mt. in R. Macedonia

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This paper provides an updated species list on the occurrence of Coleoptera fauna of Skopje city. The research was carried out during summer 2016, with a monthly dynamic. The material was collected

from five localities, by using pitfall traps placed along a transect. As a result, 75 species of Coleopterans belonging to 20 families, 29 subfamilies and 37 genera were registered, with highest species richness (42) of ground-beetle representatives (Carabidae).

Keywords: taxonomy, Coleoptera, Skopje Valley, Vodno Mt.

Medicinal fungi in Macedonia and its use

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Throughout the years, fungi made an enormous progress in medicine, despite still being a unsearched territory in Macedonia. The medicinal properties are compiled on the basis of information gathered by literature reviews. More than 50 medicinal fungi have been recorded in Macedonia. Using literature review it was established that different species have different medicinal functions and uses, such as antitumor, antibacterial, antibiotic, antiviral, cytostatic, antihyperglycemic, anticoagulant, antioxidant, antifungal and antimicrobial ones. The substrates of the fungus are lignicolous (33) and terricolous(33). According to the literature, it has been showed that medicinal fungi play a very important role in medicine.

Keywords: Medicine fungus, Macedonia, Antitumor

Diversity of macrofungi in beech forests in Ovčar-Kablar Gorge

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Scientific Research Society of Biology and Ecology Students "Josif Pančić", Novi Sad

Ovčar-Kablar Gorge is located in western part of the Republic of Serbia where beech (*Fagus sylvatica*) forests are dominant, which have a very important role in fungal diversity. Due to anthropological impacts these forests are being faced with degradation which is reflected on biodiversity. The aim of this research was to gather data on diversity of macro fungi in beech forests of Ovčar-Kablar Gorge in order to have better insight into further research regarding forest conservation. Material collected during fieldwork was identified by macroscopic characteristics of macro fungi. Research was carried out occasionally from 2013 to 2016, during which 8 different sites with beech forests were visited. Total number of recorded species on these sites was 178, out of which 57 were mycorrhizal, 117 saprotrophs, 1 saproparasitic and 3 parasitic. By the Legislation of the Republic of Serbia there were 4 strictly protected species and 3 protected species. Under the Regulation on putting the use and wildlife under control there were 2 species. Preliminary results show significant diversity of macromycetes, however it is necessary to continue research on fungal diversity of the researched area in order to obtain more reliable data which can be valuable in putting habitats under protection.

Keywords: checklist, *Fagus sylvatica*, macromycetes, protected species

Seasonal variation of the arbutin content in wild growing population of *Arctostaphylos uva-ursi* (L.) Spreng from Korab Mountain

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Arctostaphylos uva-ursi (L.) Spreng. from Ericaceae, also known as bearberry is a small procumbent woody groundcover shrub, widely distributed on a global level. The crude drug of *Arctostaphylos uva-ursi* consists mainly of three groups of pharmaceutically relevant compounds, arbutin being the main phenolic constituent. According to Ph.Eur. 8.0, commercial forms that are used consist of whole or cut, dried leaf of *Arctostaphylos uva-ursi* (L.) Spreng, that contains not less than 7.00% of anhydrous arbutin (Ph. Eur. 8.0., 2014). It was used for treatment of different diseases such as hydrops, lithiasis, in diabetes, for the therapy of gonorrhoea, etc. Nowadays only the use as urinary tract antiseptic and diuretic remains due to the presence of arbutin and hydroquinone. Therefore, the aim of the present study was to assess the arbutin content of bearberry during its vegetative cycle, from May to October, and to determine optimum technical maturity. The content of arbutin expressed as % of anhydrous arbutin, was determined by high performance liquid chromatography (HPLC). The obtained results showed that in September there is the highest arbutin content in the leaves of *Arctostaphylos uva-ursi* (8.75% of dry weight).

Keywords: Arbutin, *Arctostaphylos uva-ursi*, HPLC, seasonal variations, technical maturity.

Succession of necrophagous fauna on pig and rat carrion in suburban area in Skopje (R. Macedonia)

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In this study, diversity of necrophagous insects, successional patterns and their potential as forensic indicators were examined. The experiment was conducted in suburban area near the park Gazi Baba (Skopje, R. Macedonia) from autumn 2015 to spring 2016. Three laboratory rats (*Rattus norvegicus wistar*; Berkenhout, 1769) and one pig head (*Sus scrofa*, Linneus) were used in the experiment. Coleoptera and Diptera adults and larvae were collected both by hand and with pitfall traps placed under and around the carcass. Temperature and rainfall were measured as factors influencing the stages of carcass decay, insect activity and abundance. The stages of decomposition recognised in this study follow Reed's classification: fresh, bloated, decay and dry stage. Data on appearance time and

length of the presence period in the particular season were recorded for forensically useful taxa. The results show that Diptera, accompanied with rove beetles (Staphylinidae) were dominant representatives on carcasses during autumn - winter period. Higher diversity of beetles, mostly Silphidae, Histeridae, Nitidulidae, Dermestidae (Coleoptera) was recorded in spring season. Community of necrophagous insects was found to be significantly affected by seasonal change. Thus, general season model of insect succession on carrion are proposed for suburban area in R. Macedonia. Implications for methods of PMI estimation (particularly the succession-based method) are also discussed.

Keywords: Forensic entomology, Diptera, Coleoptera, decomposition, insect succession, R. Macedonia

Diversity and ecology of *Diatoma* sensu lato species from Macedonia

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The genus *Diatoma* sensu lato is characterized by presence of internal ribs, one or two rimoportulae near the valve apex, simple unoccluded areolae and absence of raphe system. Due to its life cycle, there is large morphological variability especially in the respect to the valve length. Such variability resulted with description of more than 350 infraspecific taxa. It is widely spread in freshwater and might be dominant in the algal assemblages. Several species are considered as eutrophication tolerant and can be found in many rivers, streams, lakes and ponds.

Detailed observations of *Diatoma* sensu lato from Macedonia have been started. The analyses resulted in reestablishing the genus *Odontidium*. Additionally, more than 15 different *Diatoma* sensu stricto taxa and at least seven *Odontidium* taxa were recorded. *Diatoma* sensu stricto might be separated into three species complexes. Most diverse is *Diatoma ehrenbergii* complex with 10 recorded taxa. Large diversity of this complex was observed in Ohrid and Prespa. Significant differences in the valve morphology between populations from oligotrophic, mountain habitats and eutrophicated habitats were noted. Such differences in previous studies were considered as ecological adaptation, but our observations show that populations have stable morphological characters that can be used for their separation. At least one unknown species was observed during this study. It is characterized by small variation in the valve size that might indicate the absence of sexual reproduction. Such phenomenon is present in a few araphid diatoms (e.g. *Meridion*), but not recorded in *Diatoma*.

The second and third species complexes *D. vulgaris* and *D. tenuis* are characterized by smaller species diversity with four and two species respectively. Both species complexes are very frequent in meso to eutrophic water bodies. Although *D. vulgaris* is considered as polymorphic species, our observations show that populations from various habitats have clear morphological differences that support their separation on species level.

Finally, the molecular analyses have been recently started. Three genes will be used for phylogenetic analyses. Two main hypothesis need to be tested: Monophyly of *D. ehrenbergii* complex from Lake Ohrid and relation of morphological and molecular markers used for separation of the species.

Keywords: Diatoma, diatoms, diversity, ecology, Macedonia

Diversity and conservation status of diatoms in the Berovo region

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The diatom flora in the region of Berovo is characterized by relatively low diversity with more than 150 species recorded. Such a low diversity is a consequence of few factors: 1) uniform geology and therefore low diversity of habitats; 2) narrow altitudinal range (absence of typical alpine habitats); 3) human impact and 4) presence of “extreme” habitats. One of the characteristics of the Maleshevo region is presence of the so called “extreme” habitats such as typical Sphagnum peat-bogs with very low pH (<4) and mineral springs. These habitats usually are inhabited by few specialized species, mostly belonging to the genera *Encyonema* and *Eunotia*. Absence of typical alpine habitats resulted with a low diversity of the genera *Eunotia*, *Stauroneis*, *Gomphonema*, and *Pinnularia*. Nevertheless, the latter genera show the greatest diversity in this region, with observed 13 and 20 species respectively.

Overall, the recorded diatom flora is consisted mainly by cosmopolitan taxa which are tolerant to higher level of eutrophication. Most diverse diatom flora was observed in the regions of Chengino Kale and Klepalo. Three species that are considered as rare in the European flora (*Caloneis aerophila*, *Navicula medioconvexa* and *Stauroneis prominula*) and few endangered species as well (*Psammodium rossii*, *Achnantidium gracillimum*, *Amphora inariensis*, *Cavinula pseudoscutiformis*, *Pseudostaurosira constricta* and *Surirella robusta*) were observed, mostly in these localities. In addition, several species, identified here as *Achnanthes* aff. *pusila*, *Adlafia* sp. *Caloneis* sp. aff. *aemula*, *Caloneis* sp. cf. *bacillum*, *Navicula* sp. aff. *recens*, possess specific characters that separate them from the most similar taxa and need more detailed observations for establishing their taxonomic status.

Contribution to the Knowledge of Ascomycetes (Fungi) in Macedonia

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This work presents the results of the research on distribution and ecology of sac fungi (Ascomycetes) in the Republic of Macedonia. The aim of this work is to provide a more comprehensive review of this fungi in Macedonia. The main part of ascocarps samplings were performed in years 2014-2016 during the spring period, mainly in March and April, covering 23 localities in various habitats, such as: deciduous and mixed forests, meadows, parks, yards and old grape vine plantations. The identification of fungi was carried out in the Mycological Laboratory, at the Faculty of Natural Science in Skopje using the appropriate literature and the following reagents: KOH, Melzer's and Congo Red. The analyzed exsiccates are deposited at Macedonian Collection of Fungi (MCF) in Skopje. During the research period, 39 species have been registered, two of them are varieties. The species belong to 18 genera: *Cyathipodia*, *Daldinia*, *Disciotis*, *Dumontinia*, *Geopora*, *Helvella*, *Microstoma*, *Morchella*, *Patiellaria*, *Peziza*, *Ptychoverpa*, *Rutstroemia*, *Sarcoscypha*, *Sarcosphaera*, *Tarzetta*, *Urnula*, *Verpa*,

Xylaria and 10 families. 31 of those are terricolous, and 8 are lignicolous. The following 8 species represent new data for Macedonian mycobiota: *Cyanthipodia villosa*, *Dumontinia tuberosa*, *Helvella macropus*, *Helvella solitaria*, *Morchella steppicola*, *Patellaria atrata*, *Peziza praetervisa* and *Sarcoscypha austriaca*. Five species are part of the Macedonian Red List of Fungi (*Disciotis venosa*, *Microstoma protractum*, *Morchella elata*, *Urnula craterium* and *Verpa conica*).

Keywords: sac fungi, diversity, new records.

Aerobic treatment of wastewater using granular microorganisms

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It is of utmost importance to provide purification of waste water that we have been constantly polluting. With this purpose waste water treatment plants are being built. The main principle of the wastewater treatment plants operation is to remove the soluble chemical compounds from the treated wastewaters so that acceptably low concentration level of pollutants has been reached. The removal of biological nutrients is carried out by using granular microorganisms that successfully remove the nitrogen and phosphorus as main pollutants of the wastewater.

Laboratory aerobic treatment of wastewater was carried based on analysis of a sample of communal wastewater (from Kadino, Ilinden municipality) using only pure granular culture and of communal wastewater from Aerodrom municipality (drain channel of communal wastewater of village Novo Lisiche) using not only pure granular culture but also pure culture of *Pseudomonas putida*. The laboratory aerobic treatment has been implemented using various methods such as: defining of volume index (Vi), biomass, Chemical Oxygen Demand (COD), NO_3^- and NH_4^+ concentration, turbidity and pH (concentration of hydrogen ions).

According to the laboratory aerobic treatment of wastewater, the best efficiency results of the volume index, turbidity reduction and biomass efficiency have been achieved when the total granular volume was 10%. Also the best efficiency for NH_4^+ concentration reduction and the biggest accumulation of NO_3^- has been identified when the total granular volume was 10%. The highest reduction of organic pollution has been noticed in cases of total granular volume was 6%. The granules have demonstrated the highest level of buffering ability in cases of total granular volume was 8%. According to this experiment, the granular microorganisms can be successfully used for the process of purification of the wastewaters.

Keywords: wastewater treatment plant, pollutants, granular culture, biomass.

Algal diversity of River Belica

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The research area of the scientific paper is a river in Republic of Macedonia, that flows through a rural area and enters the River Treska. This area is interesting because there are a lot of Balkan endemites from the floral and fungal field, but it has never been researched in the field of algae. Because of that, the subject of the investigation presented in the paper is diversity of the algae organisms in river Belica. The research is based on five different chosen locations, in order to get representative results about the algal diversity. The collected samples from the river were transferred in a laboratory, where were developed native and fixed microscopical preparations to determinate the algal species. While comparing microscopical samples with the appropriate literature, it is noticed that there are a lot of algae (cyanobacteria and green algae for examples), that don't have pictures and drawings of their characteristics. This motivated us to richen and upgrade the algal literature of this region with images and drawings of the algae with their specific details. With the results of this research we also aim to motivate further work on algal material in our country and Balkan Peninsula.

Keywords: microscopical preparations, algal characteristics, cyanobacteria, green algae, algal literature.

Diatom diversity in the region of Mariovo

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The region of Mariovo has rarely been a focus of diatom floristic studies. During July 2012, on a research camp of the Biology Students Research Society, 87 samples from diverse freshwater habitats were collected. The habitats vary in type and altitude, including small springs, streams, rivers, peat bogs, as well as wet rocks and ponds of various sizes. In total 262 diatom taxa were observed, 236 species and 26 infraspecific taxa. The most diverse diatom genera are *Gomphonema* (24), *Pinnularia* (24), *Navicula* (18) and *Sellaphora* (11). Based on the existing data on distribution and threat of diatoms species, several recorded taxa are considered as endangered or extremely rare, such as *Cocconeis neodiminuta* Krammer, *G. parvulus* (Lange-Bertalot & Reichardt) Lange-Bertalot & Reichardt, *Frustulia spicula* Amossé, *P. streptoraphe* Cleve, *Psammothidium rechtensis* (Leclercq) Lange-Bertalot. For a number of taxa (marked as "sp.", "aff.", "cf.") complete identification was not possible using the existing literature. These taxa require more detailed taxonomic treatment to establish their identity. Till now, two new species were described, *G. ristovskae* Levkov & Tofilovska and *G. mariovense* Levkov & Tofilovska. Interestingly, in the material collected from some of the streams and rivers, two species, *Tertiarius*

jurijlii Ognjanova–Rumenova et al. and *T. mariovensis* Ognjanova–Rumenova et al., described from sediments of Pliocene age from the Mariovo Basin were observed. The genus *Tertiarius* comprises species described only from fossil deposits, from the Miocene and Pliocene periods, so the presence of this individuals is most probably due to the flushing of sediments. The most significant diversity was observed in the area of Melnicki Škrki where the highest number of taxa was recorded. This is possible due to the existence of a variety of habitats present in this locality. The diversity of Mariovo diatom flora as observed herein is far from complete.

Keywords: diatoms, Mariovo, Melnicki Škrki, new species, recent flora, *Tertiarius*

Rove beetle *Dinothenarus flavocephalus* (Goeze, 1777) (Coleoptera: Staphylinidae) new forensic indicator of season of death

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Rove beetles (Staphylinidae) are common predators found on corpses. The subfamily Staphylininae includes species of the largest forensic importance. In order to evaluate usefulness of Staphylininae as indicators of season of death, a rat carrion experiment was conducted from autumn 2015 to spring 2016 in suburban area near the Gazi Baba Park (Skopje, Macedonia). During the study on entomofaunal succession on decaying rat carrion, seven specimens of rove beetle *Dinothenarus flavocephalus* (Goeze, 1777) were collected. This species is reported for the first time for the Republic of Macedonia, extending its known distribution in the Middle East and in several European countries. Additionally, clear seasonality in this species, occurring only in autumn - winter seasons was confirmed. No specimens were registered during spring season. Results indicate that *D. flavocephalus* is good candidate for new forensic indicator of season of death. To assist in the recognition and the use of this species in forensic entomology, diagnosis and illustrations of morphological features useful for identification of adults are also provided.

Keywords: Rove beetles, *Dinothenarus flavocephalus*, first record, Republic of Macedonia, forensic indicator

Conservation-important butterflies in an under-researched part of Macedonia, Plachkovitsa and Maleshevo Mountains

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Being of high conservation interest, butterflies are one of the most studied groups of insects in Europe. With nearly 10% of endangered species on European level, each small contribution of the distribution status of the butterflies represents a significant input in further conservation efforts. Although the butterflies are fairly studied insect group in the Republic of Macedonia, more knowledge is needed on the presence and distribution of the conservation-important species. Our study focuses on faunistic research of butterflies on Maleshevo and Plachkovitsa Mountains located in the eastern part of the country. Each of these mountain ranges offers diverse habitats that could support healthy populations and rich diversity of butterflies. Therefore, by using random sampling as a research method, we conducted a field research on these mountains in July 2014 and 2015. A wide array of localities and habitats have been researched in which a total of 82 species were registered. Maleshevo and Plachkovitsa Mountains support 40% of the total number of butterfly species in Macedonia. During our research, we have recorded a total of six butterflies of high conservation interest which demonstrates the importance of this region for butterfly species richness. Future studies should be focus on abundance estimates and habitat availability of targeted species that are of high conservation priorities.

Keywords: Maleshevo and Plachkovitsa Mountains, butterfly diversity, field research, butterfly conservation, conservation - important species

Great Cormorant colony at the island of Golem Grad, Greater Prespa Lake: breeding numbers and nest tree selection

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The only known breeding colony of the Great Cormorant *Phalacrocorax carbo* in Macedonia is found on the island of Golem Grad, at Lake Prespa. Its size has not been estimated since 1979. To overcome this situation, a nest count was carried out after the breeding season of 2016, covering the entire island. Five teams of two people tried to locate all of the Cormorant nests, and to estimate how many of them have been used in the respective season. Such estimation was straightforward for most of the nests, but an estimated 15% error might be possible, as sometimes we were not able to make a conclusive decision. In total, there were 1487 nests situated on 500 trees. It was estimated that 1237

nests were active and 250 were not used in 2016. The average number of active nests per tree was 2.47 (ranging 1 - 10). Cormorants mostly nested on the Greek Juniper *Juniperus excelsa* trees (1461 nests), which are dominant tree species on the Island. The rest were distributed among *Celtis sp.* (24), *Salix sp.* (1) and *Prunus sp.* (1). Regular monitoring of the colony will be required to establish its dynamics and breeding parameters, as well as its effect on the Greek Juniper forest on the Island.

Key words: census, Greek Juniper, *Juniperus excelsa*, *Phalacrocorax carbo*, population estimation

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