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Editors' remarks

The presentation of the material in this document do not imply the expression of any opinion whatsoever on the part of the Croatian association of freshwater ecologists or of other institutions involved in the organization of the Symposium. The views expressed in this volume are those of the authors of each abstract, while editors preformed technical editing and equalized the form of abstracts. The quality of the English language is the responsibility of the authors, as no substantial language editing of submitted texts was done.

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Preface

A handful of enthusiastic biologists established Croatian Association of Freshwater Ecologists in February 2014 and since then we are working on inter-institutional cooperation in Croatia and abroad. We soon organized the First Symposium of Freshwater Biology, which was held in Zagreb on February 15th, 2015 in order to present current information on freshwater ecosystems in Croatia and beyond, and gather students, scientists, experts and enthusiasts all in one place to communicate and collaborate. Since the First Symposium proved to be very successful and interesting, we decided to continue with the symposiums every two years. Our Second Symposium of Freshwater Biology was held in Zagreb on February 17th 2017. The Second Symposium was even more successful with more than 100 participants. Our Third Symposium of Freshwater Biology is set for February 15th, 2019. All our three symposiums were organized in cooperation with the Department of Biology, Faculty of Science, University of Zagreb. We are happy that this event brings together a large number of biologists of all ages, including biology students that present their own research under the supervision of their mentors. In this publication, which includes all abstracts presented during the symposium, you will see a diverse and interesting current expert and scientific topics associated with all aspects of freshwater biology. We hope that you will find some new ideas, challenges, opportunities for cooperation, gaining new colleagues and friends, or at least get enthusiasm for future work, so you could actively engage into research, education and protection of freshwater ecosystems.
We thank all the participants who have recognized our original idea, and made this event possible.

See you in year 2021.

President of the 3rdSymposium of Freshwater Biology

Marija Ivković

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Dr Thibault Datry works at the National Research Institute of Science and Technology for Environment and Agriculture (IRSTEA) in Lyon, France. His research focuses on the ecology of rivers and hyporheic zones, with particular emphasis on intermittent rivers. He has published more than 120 peer-reviewed papers on stream and groundwater ecology and co-edited the first book on intermittent rivers and ephemeral streams. He led the international synthesis project IRBAS (irbas.cesab.org/) and chairs the SMIRES Cost Action (www.smires.eu) on the science and management of these ecosystems.

Intermittent rivers and ephemeral streams: a challenge for freshwater science

For several centuries, freshwater research has focused on perennial rivers and streams which flow all year round. However, those which don't, because they cease to flow or dry up completely at some stage in space and time, (hereafter intermittent rivers and ephemeral streams, IRES), are a very recent addition to the field, although they comprise more than half of the global river network and dominate in many areas, including Mediterranean regions. Concepts that have guided studies of biological communities and biogeochemical fluxes, and material exchange between rivers and the atmosphere, land, ocean and groundwater presume perennial flow and continuous hydrological connectivity. In IRES, the loss of hydrological continuity affects virtually all ecological processes, including the evolution, dispersal and habitat selection of aquatic and terrestrial species.

Owing to the recent and intense research efforts on IRES ecology, our views of the roles that rivers play in maintaining biodiversity, vital ecological processes, and controlling material fluxes is expanding exponentially. Most river networks are composed of IRES, whose prevalence is increasing due to global change. IRES are dynamic mosaics of aquatic and terrestrial habitats, each hosting rich and unique biotic communities, some contributing substantially to key ecosystem functions such as the decomposition of the terrestrial litter. But IRES are not only ecosystems on which concepts developed for perennial systems have been tested; they are today model arenas to progress general ecology. For example, community ecology has progressed rapidly owing to the recognition that local communities are shaped by both local (biotic interactions, responses to abiotic environmental conditions) and regional (dispersal of species in a region) processes. However, research on freshwater metacommunities offered ambiguous explanations for the relative importance of local vs regional processes and most developments on metacommunities have considered both biological communities and their habitats as relatively stable, limiting our understanding of the temporal dynamics. IRES provide especially suitable arenas for examining metacommunity organisation in an extremely dynamic setting, where aquatic and terrestrial communities alternate periodically locally and coexist in a drainage network. Metacommunity organisation in the wet and dry phases of IRES are closely intertwined and affect the succession of each other. However, conceptual developments are needed to incorporate such temporal variability. More globally, such ecological setting offers a unique

opportunity to bring together the concepts, questions, approaches and methodologies from lotic, lentic, and terrestrial ecology, to increase our knowledge and manage these rivers more effectively. Such approaches will not only help advance the science of IRES, they will also help to develop new theories and models, and design management plans and policies for all dynamic ecosystems facing global change.

In this talk, I will illustrate these ideas through data collected and ideas shared in PhD and postdoctoral projects in my department and in global collaborative projects, such as the 1000 Intermittent River Project and the SMIRES Cost Action. I'll also provide a roadmap to stimulate further developments of IRES research and other highly dynamic and fascinating ecosystems.

Tomáš Hauer

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Dr Tomáš Hauer works at the University of South Bohemia, České Budějovice, and Institute of Botany AS CR, Třeboň, Czech Republic. His research is focused mainly on diversity and taxonomy of Cyanobacteria from wide range of habitats including natural aquatic, terrestrial as well as industrial. He is a co-leader of the Determination Course of Freshwater and Terrestrial Cyanobacterial held regularly at the University of South Bohemia specialists from the field. As a side-project he collaborates on algal diversity monitoring in nature protected areas.

How to navigate through current cyanobacterial taxonomy

Cyanobacteria represent an important group of organisms in both aquatic and terrestrial ecosystems. Their diversity has been thoroughly studied since 19th century and with increased application of molecular methods we are facing an avalanche of new taxa. Almost every month, numerous new genera and species are being described. Simultaneously, but in much lesser extent, revisions of formerly described genera or higher taxonomic units are performed bringing in certain cases unexpected results. Besides newly described species and genera, many changes take place in classification to families. Even though results of revisions and new taxa delimitations may be confusing for users or may cause inconvenience in routine analyses, new findings should be reflected in practice as well as in other research as soon as possible. Unfortunately, the findings and consecutive results may be biased by several factors which influence their reliability, and which may be less obvious for specialists from different fields outside of the cyanobacterial taxonomy. To main of these factors belong a substantial number of sequence names incorrect for several reasons which are stored in main public molecular data repositories (i.e. NCBI GenBank, EMBL ENA, DDBJ) and their use as the sole determination tool without including other information and data such as morphology or ecology. Next factors are the lack of reliable reference data (i.e. molecular data on type species) for many genera or low experience of authors of certain works with nomenclature rules. Last but not least two sets of nomenclature rules, i.e. “Botanical code” (ICN) and “Bacteriological Code” (ICNP), which are not fully compatible with each other, are applied on Cyanobacteria. All above mentioned points affect results of both “16S rRNA gene only” as well as “genomic” taxonomic works, and consequently also the results of other research that relies on taxonomy or comparison of the newly obtained sequences with data available, especially in case of environmental sequencing. The present talk will deal with examples of recent findings in taxonomy of certain important genera (e.g. *Oscillatoria, Arthrospira*), and in detail with above mentioned factors to help non- taxonomists to navigate through the new findings and to help them recognize important and relevant taxonomic information among all the data available. As one of the tools allowing easier orientation in the given topic, the new version of the CyanoDB.cz database will be introduced.

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Protist assemblages associated with the endemic subterranean bivalve *Congeria jalzici* Morton & Bilandžija, 2013 (Bivalvia: Dreissenidae)

Biodiversity and ecology of organisms living in karst subterranean habitats of Dinaric arc are poorly known. Data gaps are especially pronounced regarding species interactions and habitat variability. The main goal of this research was to enhance our knowledge of environmental conditions and communities present in habitats of Natura 2000 endemic species of subterranean bivalve *Congeria jalzici,* in order to better understand its ecology. Preliminary research of protists and other present microorganisms in eight speleological objects of wider area of Lipovo polje (Lika, Croatia) was performed during 2016 and 2018. It included microhabitats characterized by complete darkness. Biodiversity and abundance of protozoa was recorded, while presence of other microorganisms was recorded only at the higher taxonomical level. Focus was given to hygropetric habitats, which are of crucial importance for *C. jalzici* during the months characterized with hydrological minimums. Protozoa assemblages from hygropetric habitats in three speleological objects inhabited by *C. jalzici* formed separate clusters compared to other five caves where *C. jalzici* was not found. Especially diverse was polyphyletic group testate amoebas with 26 species, including some very rare species. Algae and cyanobacteria, transported probably by floods, were detected in some caves indicating specific hydrological connectivity. During this research, for the first time epibiotic ciliates on *C. jalzici* were registered and histologically confirmed. These findings show that assemblages of protists and other microorganisms in subterranean habitats, even though often neglected, should be considered in context of *Congeria* ecology especially because of their potential as bioindicators of environmental conditions.

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Larval chironomid community of Prošće and Kozjak Lakes, Plitvice Lakes National Park

Chironomid community is usually the most abundant and diverse of all benthic communities. Plitvice Lakes are the oldest Croatian National Park, which consists of 16 barrage lakes. Littoral zones of two of the biggest lakes, Prošće and Kozjak were sampled on one occasion, in July and September 2018, respectively. At each lake, five sampling sites were chosen. Each sampling site covered the length of 25 m lakeshore and a distance of 10 m towards the open water or alternatively, to the point at which the water depth exceeded 1 m. Four water-depth classes were defined: 0–0.25 m, 0.25–0.5 m, 0.5–0.75 m and 0.75–1 m and microhabitat types were also regarded in sampling procedure. At each sampling site, ten samples were collected using a benthos hand net. In total 12 895 individuals belonging to 25 taxa were collected. The most abundant genus in the Kozjak Lake was *Cladotanytarsus* and in the Prošće Lake a predaceous genus *Conchapelopia*. The abundance, number of taxa, diversity indices and evenness index were all slightly higher in the Kozjak Lake as well as water temperature. On the other hand, oxygen concentration, water conductivity and pH were higher in the Prošće Lake. The differences in chironomid composition between lakes are most likely due to differences in environmental conditions between lakes.

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Update on signal crayfish, *Pacifastacus leniusculus* (Dana, 1852), range expansion in Croatia: A 10-year report

The signal crayfish (*Pacifastacus leniusculus*) is the most successful crayfish invader in Europe. In Croatia it was firstly recorded in the Mura-Drava river basin in 2008, as a result of natural dispersal by Slovenian populations. In 2012 it was recorded in the Korana River in the continental part of Croatia, where it was illegally introduced. In both rivers, a high dispersal rate was observed. The aim of this study was to explore the range expansion of the signal crayfish 10 years after its first record in Croatia. We collected literature data and conducted field studies in summer 2018 along the Korana and Drava rivers. In the Korana River, we observed an increase in the range of *P. leniusculus* by 5 km in 2 years. The invasion range now stretches along a total of 30 km. Dispersal rates were higher in the upstream direction, potentially due to differences in competitive pressure from native crayfish. Abundance of the native narrow clawed crayfish (*Astacus leptodactylus*) was six times lower at the upstream invasion front than downstream. In the centre of the *P. leniusculus* invasion range in the Korana river no native crayfish were captured, suggesting a displacement of the narrow clawed crayfish by the invasive signal crayfish. In the Drava River, the extent of the newly recorded invasion front corresponds to projections from an earlier study that estimated signal crayfish dispersal over 10 years. Our new records suggest that signal crayfish dispersal rates in this river have remained among the highest in Europe.

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Macrozoobenthos community in fast flowing streams of Slavonian mountains

The Pannonian ecoregion of Croatia is characterized not only by lowland features, but also with a mountain range consisting of Psunj, Papuk and Krndija mountains (west and north), and Požeška Gora and Dilj (south and east). As a recognition for its unique geological and biological features, Papuk was pronounced Nature Park and Geopark. However, the published material on its diversity is still scarce, especially for macroinvertebrates. Our aim is to shed some light on the diversity of the stream invertebrate fauna and their application in water quality assessment. The sampling was conducted in spring 2017 in Velika Rijeka (Krndija) and Šumetlica (Psunj) streams and in Jankovac (Papuk) stream in spring 2018. Quantitative sampling of the macrozoobenthos was performed, following the AQEM protocol (multi-habitat sampling), using 500 µm mash hand net, and physico-chemical water parameters were measured concurrently. Macroinvertebrates were isolated in the laboratory and identified to the genus/species level. Total number of identified taxa was 102, with 44 species identified. The highest biodiversity was recorded at Jankovac site, followed by Šumetlica and Velika Rijeka streams. Chironomids and oligochaetes were the most abundant in Velika Rijeka (816 and 89 Ind. / 0.0625 m^2, respectively) whilst the EPT group had the highest abundance and diversity in Jankovac stream. Aforementioned groups are used in water quality bioassessment protocols and all three streams indexed “good” ecological state. This preliminary study reveals a great diversity of macrozoobenthos community in the understudied Pannonian streams and thus serves as a good starting point for further research.

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Microhabitat preferences by aquatic invertebrates in the Zrmanja River springhead

Karstic springs are among the most complex ecotonal environments with specific transition zones based on hydromorphological properties. Nevertheless, most ecological studies of spring assemblages are based on distribution differences between crenal spheres (i.e. eucrenal vs hypocrenal). The ecological study of benthic invertebrates in the Zrmanja River springhead was focused on spring microscale in order to provide deep insights into fauna's preferences of key physical variables. During April 2016, we collected 40 subsamples in dominant microhabitats (i.e. substrates with high and low water velocity) using the hand net according to modified AQEM procedure. The springhead assemblages were generally dominated by its temporal residents, Diptera and Ephemeroptera. The prevalence temporal fauna were Chironomidae, being the most abundant at the moss in the low water velocity. According to the EPT index, the highest EPT taxa richness was recorded in moss on the macrolithal and megalithal substrates exposed to the high-velocity water flow. In the mesolithal, predatory Plecoptera (Perlidae) were positioned at the top of the food web in the low water velocity, while they were replaced by Trichoptera (Rhyacophyllidae) in the high water velocity. Hydrachnidia, as a key spring fauna, were recorded mostly at microhabitats with low water velocity within the microlithal. We confirmed Reiss and Chifflard conceptual model of eucrenal as a heterogeneous mosaic-like patches and recorded heterogeneity on multiple levels.

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Macroinvertebrate community of a temporary Mediterranean river

Temporary rivers and streams are lotic ecosystems which are characterized by partial or complete lack of surface water in dry seasons. They have great impact on hydrology of adjacent perennial water bodies. However, until recently their importance was often overlooked, which led to degradation of these habitats. In last decades, number of studies significantly increased and an initiative for development of management procedures was launched. This research was conducted along a temporary karst river Krčić, situated beneath the Dinara Mountain. Krčić is a small river which usually dries up during summer and early autumn months. It is minimally influenced by anthropogenic activities and can thus be regarded as a reference site for temporary rivers in this region. Sampling of macroinvertebrates was performed in 2016 during the flowing phase; in June and December, on the upper, middle and lower reaches. It encompassed all major microhabitat types (moss, fital, megalital, makrolital, mikrolital and xylal). At each site several physico-chemical parameters were measured. Community composition and structure were typical for temporary river communities, with Chironomidae as the most abundant insect group (14 taxa), followed by Plecoptera (4 taxa), Ephemeroptera (3 taxa) and Trichoptera (3 taxa). Generally, high diversity and evenness of the community were observed. The results of cluster analysis and MDS showed macroinvertebrate community separation by sampling seasons, not by macrohabitats. These results highlight the importance of seasonality in temporary rivers and streams, therefore it is necessary to perform seasonal monitoring at least twice a year.

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Short-term effects of natural stream discharge on the water quality trends along two urban streams

Environmental changes associated with urbanization primarily modify stream morphology, which then further affects physical (discharge), chemical (water quality) and biological (biota) status of urban streams. We investigated the short-term effects of natural stream discharge on the water quality along two streams (Bliznec, B and Veliki potok, VP) flowing through Zagreb, Croatia. The research was conducted at six study sites stretched along the two urban streams during a two-week period in October 2017. Reference sites (B1, VP1) were located in the upper stream reaches within the Medvednica Nature Park. Other sites were aligned along an increasing urban gradient within Zagreb – sites B2 and VP2 were located in the middle, and B3 and VP3 in the lower stream reaches. The following physico-chemical parameters were measured every other day over the two-week period: stream discharge, water temperature and oxygen concentration, conductivity, chemical oxygen demand (COD_{KMnO4}), and in-stream concentrations of nutrients (nitrites, nitrates, orhophosphates). Nutrients were the most variable parameters during the two-week period. Bliznec generally showed significantly higher discharge and oxygen levels compared to Veliki potok, which had significantly higher COD_{KMnO4} and nitrite concentrations. Middle and lower stream reaches had significantly higher temperature, nitrite and nitrate concentrations in comparison to the upper reaches. The data also revealed significant between-site and temporal changes of the physico-chemical parameters after a major storm event. Moreover, conductivity was found to correlate with discharge. Our findings indicate that urban streams are very dynamic ecosystems showing an increased sensitivity on environmental changes, such as anthropogenic influences and weather conditions.

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Morphometry of freshwater organisms as useful method in ecotoxicology

Ecotoxicological observations of cytological measurements were performed on the freshwater planarian *Polycelis felina* (Dalyell, 1814) and cnidarian *Hydra viridissima* (Pallas, 1766) with the aim to determine morphometrical changes of their cell structures. *P. felina* was exposed to aluminium (80 and 100 mgL^{-1} for 48 h). *H. viridissima* was exposed to cinoxacin ($1.9x10^{-3}$ and $3.8x10^{-3}$ mol/L for 72h) and chloramphenicol ($2x10^{-4}$ mol/L for 24h and $3x10^{-4}$ mol/L for 24 and 48h). Both species were exposed to herbicide norflurazon ($2x10^{-6}$ mol/L) for 3, 24 and 48h. In hydras morphometrical changes were observed in number and area of zymogen and interstitial cells, area of ectoderm and gastroderm, mesoglea thickness, size of ectodermal myoepithelial cells and diameter and area of endosymbiotic algae. In planarians morphometrical changes were observed in the size and number of neoblasts and in the size of the reticular cells, rhabdites and rhabdite layer. The results of morphometric analysis showed changed structures of different cell types, significantly correlating with an increase of toxicant concentration and longer exposure, compared with the control samples. Morphometry proved to be a powerful tool in research of ecotoxicological impact of xenobiotics upon freshwater organisms.

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Predictive ecotoxicology: are we there yet? Case study of river sediment toxicity assessment - combining empirical zebrafish embryotoxicity testing with *in silico* toxicity characterization

Quantitative chemical analyses of 428 organic contaminants (OCs) indicated the presence of 313 OCs in the sediment extracts from Sava River, Croatia. Pharmaceuticals were present in higher concentrations than pesticides thus confirming their increasing threat to freshwater ecosystems. Toxicity evaluation of the sediment extracts from four locations (Jesenice, Rugvica, Galdovo and Lukavec) using zebrafish embryotoxicity test (ZET) accompanied with semi-quantitative histopathological analyses exhibited correlation with cumulative number and concentrations of OCs at the investigated sites (10.05, 15.22, 1.25, and 9.13 µg/g respectively). Toxicity of sediment extracts and sediment was predicted using toxic unit (TU) approach and persistence, bioaccumulation and toxicity (PBT) ranking. Additionally, influential OCs and genes were identified by graph mining of the prior knowledge informed, site-specific chemical-gene interaction models. Predicted toxicity of sediment extracts (TUext) was similar to the results obtained by ZET and associated histopathology with Rugvica sediment being the most toxic, followed by Jesenice, Lukavec and Galdovo. Sediment TU (TUsed) favoured OCs with low octanolwater partition coefficients like herbicide glyphosate and antibiotics ciprofloxacin and sulfamethazine thus indicating locations containing higher concentrations of these OCs (Galdovo and Rugvica) as the most toxic. Results suggest that comprehensive *in silico* sediment toxicity predictions advocate providing equal attention to organic contaminants with either very low or very high log Kow.

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Ulva flexuosa Wulfen in fresh and brackish waters of Croatia

Genus *Ulva* Linnaeus (Ulvophyceae, Ulvales, Chlorophyta) is a cosmopolitan macroalgal genus commonly known as „sea lettuce“. It comprises species living in different aquatic environments, however represents the most frequent coastal seaweed world-wide. Nevertheless, since the mid 1800's *Ulva* is being observed in European inland waters as well. At the beginning of the 21st century, massive occurrences of *Ulva flexuosa* have been noted in European lakes, ponds and rivers, making the genus an obvious dominant element in these waterbodies. In Croatia, a number of specimens involving *Ulva* (including former *Enteromorpha*) have been recorded in inland freshwater and brackish sites of northern and middle Adriatic, casually and erroneously described as *Enteromorpha intestinalis* (Linnaeus) Nees (=*Ulva intestinalis*) and *E. salina* Kützing (=*U. prolifera*). Intrigued by the observed ingress of sea taxa towards freshwaters, we collected specimens from freshwater sites along Eastern Adriatic coast in Croatia while conducting the monitoring of macrophytes. Additionally, macrophyte vegetation surveys were performed and physico-chemical parameters were measured *in situ* or by Central Water Management Laboratory. In the period 2016-2018 *Ulva flexuosa* has been recorded in Raša, Mirna, Zrmanja, Cetina rivers, water bodies in the Neretva River Delta and in freshwater spring on the island of Krk. In these locations, salinity ranges were from 0 to 18 PSU, while species occasionally occurred in massive blooms and undoubtedly formed significant habitats in the form of rooted or floating aquatic vegetation. These findings mark the first data about occurrence of *U. flexuosa* in Croatian fresh and brackish waters.

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Impact of invasive peracarid crustaceans on benthic macroinvertebrate assemblages in Croatian large rivers – preliminary assessment

In this study we investigated longitudinal distribution changes and density of native and alien Peracarida (Amphipoda and Isopoda) in Croatian large rivers and their proportion in total density of benthic macroinvertebrate assemblages. Qualitative and quantitative benthos samples collected using hand net (25x25 cm, ⌀500 µm) in five major large rivers from 2004 to 2018 at 236 sites were used for distribution changes. Peracarid densities and proportions in macroinvertebrate assemblages were established using quantitative samples collected at 48 sites in 2015 and 2016. In a total 16 peracarid species were recorded, eleven alien and five native. *Dikerogammarus haemobaphes* was the most upstream distributed alien species in the Sava River and *D. villosus* in the Drava River. Upstream spread of *D. haemobaphes* in Sava (front at 648 rkm in 2018) was much faster during the last three years (2015-2018: 4.6 km year^{-1}), than before (2012-2015: 0.9 km year^{-1}). Front of *D. villosus* in Drava River was at 289 rkm in 2018, while its upstream spread ranged 2-22 km year^{-1} in the study period. Proportions of alien species in the total abundance of benthic assemblages were very variable (range: 0.01-90.9%, avg: 29.7%) and species with the highest densities and proportions were *Chelicorophium curvispinum* in the Drava and *C. sowinskyi* in the Sava River (>70% at some sites). Upstream sections of Sava and Drava had lower densities and proportions of invaders in macrozoobenthos than downstream sections. Future studies should assess the spread and impact of alien peracarid crustaceans on macroinvertebrate assemblages in Croatian rivers.

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Fountains – extreme habitats or source of biodiversity in the urban areas?

Within the urban areas, there can be various natural and artificial water-bodies, supporting diverse biota. City fountains are often ignored by aquatic ecologists, since they represent more extreme aquatic habitats, simultaneously having characteristics of lotic and lentic water-bodies, often treated with chemicals. However, the combination of those conditions can sustain rather specific invertebrate communities, and Chironomidae (Diptera) can be a significant part of them. Three fountains in Osijek were investigated from April to October 2016, where we sampled chironomid pupal exuviae, using a 300µm mash net and concurrently measured basic physico-chemical water parameters. In the laboratory, permanent microscopic slides were made using the "Berlese" mounting medium, to identify collected chironomid specimens. Twenty-nine taxa were recorded, belonging to two subfamilies: Orthocladiinae (15) and Chironominae (14), with all fountains supporting similar taxa richness. The highest abundance, 451 exuviae, and diversity, 10 taxa, were found mid-June, in the F3-Mačkamama fountain. The most abundant and frequent species (53.3%) was *Cricotopus sylvestris* (Fabricius, 1794). PERMANOVA analysis did not show statistically significant differences in the community structure among fountains or seasons. Additionally, according to the BIO-ENV analysis, there was no significant correlation with the water parameters; thereby the community structure was most likely driven by the natural seasonal variability, differences in life cycles, or taxa richness and other sources of re-colonization. The number of Chironomidae taxa recorded in this research shows the importance of the fountains as a source of biodiversity in the urban areas, but also validates chironomids as a very adaptable insect family.

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Effects of pharmaceutical pollutants on physiology of the invasive signal crayfish, *Pacifastacus leniusculus* (Dana, 1852)

Pharmaceutical pollution in freshwaters can affect behavior and physiology of freshwater organisms since most pharmaceuticals are designed to act quickly in human body and leave the body without degrading, so they enter the freshwaters still pharmacologically active. Recent studies have demonstrated that pharmaceuticals can exhibit effects on behavior and physiology of freshwater organisms in environmentally relevant concentrations. This study explores whether pharmaceuticals commonly present along the Croatian section of the Sava River (antidepressants Citalopram and Nortriptyline, and illicit drug Amphetamine) could have an effect on physiology of the most successful crustacean invader of European freshwaters, Signal crayfish (*Pacifastacus leniusculus*). We exposed individuals to environmentally relevant (low), or therapeutic (high) concentration of Citalopram, Nortriptyline or Amphetamine dissolved in ambient water for 7 days. Since antidepressants act via alterations in serotogenic metabolism and receptors and since in crayfish serotonin mediates the release of cHH, hormone that controls hemolymph glucose concentration, we chose to measure the following physiological endpoints after 7-day exposure: concentration of serotonin in the brain, concentration of glucose in hemolymph, as well as markers of oxidative stress. We compared the results obtained for exposed individuals to their respective control groups. We did not observe any influence of examined pharmaceuticals on serotonin levels, and in concordance to our expectations exposure to tested concentrations did not exert any oxidative stress upon individuals. We also did not observe significant effects on glucose levels except for Amphetamine where glucose levels were lower than in control group at both concentrations.

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A new diatom species from Lake Crveno jezero, Croatia (*Gomphosphenia plenkoviciae* sp. nov.)

The genus *Gomphosphenia* Lange-Bertalot was separated from *Gomphonema* Ehrenberg and when established, it was considered a species limited genus with 17 taxa including species and varieties. None of the species from the genus *Gomphosphenia* have yet been recorded in Croatia and for the first time was observed in Lake Crveno jezero (Red Lake). The lake is one of the largest karstic sinkhole lake in Europe, situated in the town Imotski in Croatia, near the border with Bosnia and Hercegovina. Diatom flora of this lake was unknown due to difficulties in collecting samples. Sampling of phytobenthos was conducted as part of systematical natural sciences research organized during the International speleodiving expedition “Crveno jezero 2017”. Observations revealed the existence of a new species, *Gomphosphenia plenkoviciae* Gligora Udovič & Žutinić sp. nov. *Gomphosphenia plenkoviciae* possesses typical morphological and ultrastructural characteristics of genus *Gomphosphenia*. It is characterized by having narrowly clavate to linear valves, which are slightly heteropolar in larger specimens to clavate in smaller specimens, thus differing from the closest taxon by the valve outline and shape of the central area.

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How well existing Natura 2000 sites protect threatened stone crayfish in croatia?

The stone crayfish *Austropotamobius torrentium* (Schrank, 1803) is one of five native European freshwater crayfish species distributed in Croatia, mainly in water bodies of the Black Sea drainage with a few populations in the Adriatic Sea drainage. The number of stone crayfish populations has been decreasing as a result of anthropogenic pressure on their habitats, climate change and spreading of invasive crayfish species and their pathogens. Due to severe declines in numbers of populations in the last decades, stone crayfish is considered threatened species and is protected by both national and international laws. The Natura 2000 network represents the most important conservation effort on the European level. In Croatia it includes 25 sites designated for *A. torrentium* that were defined based on historical data and extensive research conducted between 2008 and 2010. The aim of this research was to assess the effectiveness of Natura 2000 sites in preserving stone crayfish diversity. Therefore, we performed gap analysis, a GIS-based approach that overlays species distribution data onto a map of designated Natura 2000 sites. Overall, our results showed that existing Natura 2000 network covered relatively well species range; currently designated sites harbour 72.5 % recorded *A. torrentium* populations in Croatia. Future conservation planning efforts, and possible expansion of Natura 2000, should be focused onto newly discovered *A. torrentium* populations that potentially represent undescribed divergent evolutionary lineages.

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Morphological diversity and distribution of *Eleocharis* taxa (Cyperaceae) in Croatia

The genus *Eleocharis* (Cyperaceae) has a circumboreal distribution and consists of about 200 species which are found in marshes, wetlands and other wet habitats. The taxa of this genus were never systematically studied in Croatian flora. Based on morphological characters a revision of roughly 300 herbarium specimens deposited in four Croatian herbaria (CNHM, ZA, ZAGR and ZAHO) was performed. The characters that are used for revision are: plant habit, uppermost leaf-sheath shape, inflorescence and spikelet shape, glume shape and length, fruit (nut) shape and length, stylopodium shape, and number of perianth bristles. The results of this revision revealed the occurrence of nine *Eleocharis* taxa in Croatian flora: *E. acicularis*, *E. carniolica*, *E. mamillata*, *E. mamillata* subsp. *austriaca*, *E. ovata*, *E. palustris*, *E. parvula*, *E. quinqueflora* and *E. uniglumis*. For each taxon the distribution map is provided and commented. Quite problematic and morphologically very similar are taxa of *E. palustris* complex (*E. mamillata*, *E. mamillata* subsp. *austriaca*, *E. palustris* and *E. uniglumis*). Their distribution and occurrence is still poorly known, expect for *E. palustris* that is wider distributed in Croatian flora. The occurrence of *E. parvula* is questionable in Croatia. The habitats of *Eleocharis* taxa are under strong human influence because of vegetation succession and draining of wet habitats, as well as abandonment of traditional agriculture on wet fields and meadows, and actual climate warming changes.

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Analysis of the re-established macroinvertebrate community in restored habitats

Krka National park is among the unique areas in the world valuable aesthetically, economically and biologically. Despite all the actions that a National Park level of protection ensures, uncontrollable spreading of an invasive plant species *Ailanthus altissima* damaged a number of habitats in Krka`s tufa barriers. Due to its overgrowth, several streams with their active tufa-depositing habitats were harmed and ultimately dried. Consequently, macroinvertebrate community disappeared causing changes in ecological features of the area and leaving a negative effect on species richness and food webs. Recovery of the area started with the removal of all the invasive trees. Once the trees were removed, regeneration of the 3 channels started. Slowly, the macroinvertebrates started recolonizing the habitats. Our goal was to determine the time required for the restored community to become comparable to the control one in the undamaged habitats with constant flow. Benthos samples were taken once a month at five locations using a core-sampler up to 3 cm depth. Simultaneously, physico-chemical properties of water were measured. Altogether, 16 macroinvertebrate taxa were isolated, among which non-biting midges were the most abundant, followed by blackflies, outnumbering the mayflies and amphipods which appeared only in 2 out of 5 sites. Although originally abundantly present at the control site, beetles were very rarely found in the rewetted channels.

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DNA barcoding of Croatian biodiversity: riffle beetles (Insecta: Coleoptera: Elmidae)

Riffle beetles are widely distributed aquatic insects inhabiting springs, rivers and streams. They are rather small sized beetles with low dispersal ability, and they are often used as bioindicators in water quality assessment. Nowadays the use of integrative taxonomy in species identification is fundamental, especially in cases of closely related species. So far, only few studies were focused on DNA barcoding of European Elmidae, as the samples from only seven European countries are included in the Barcode of Life Database. The aim of this study was to conduct: i) identification and DNA barcoding of elmids held in museum collections; ii) sampling and barcoding of additional specimens throughout Croatia; iii) forming of comparative beetles collection of Croatian Elmidae. In total, 130 specimens were included in the study representing 18 out of 24 recorded species in Croatia up to date. Identified and DNA barcoded specimens were photographed and stored, together with male genitalia collection that was formed within the Water beetle collection at Croatian Natural History Museum. In comparison with European Elmidae our results indicate the existence of potentially rare and/or endemic species in Croatia, with significant genetic differentiation of certain populations. Future study will include sampling of species that were not yet analyzed, with special emphasis on rare and/or endangered species such as *Stenelmis consobrina* Dufour, 1835 or *Potamophilus acuminatus* (Fabricius, 1792). Additionally, this study provides the basis for future research of riffle beetles in Croatia, as well as it contributes to the protection and conservation of Croatian freshwater biodiversity.

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Factors influencing littoral chironomid assemblages in the reservoirs of the Dinaric karst

In this study, littoral chironomid assemblages were investigated at 11 reservoirs in the Dinaric Western Balkan ecoregion. Reservoirs are complex systems that represent a transition between lakes and rivers and it has been used for irrigation, flood control and electricity generation. Chironomids were the most abundant (aquatic) insect group in the littoral macrozoobenthic community. Larvae were sampled during the summer months of 2016. At each sampling site, ten samples were collected using a benthos hand net. A total of 36 taxa were recorded, belonging to four subfamilies: Chironominae (Chironomini and Tanytarsini), Orthocladiinae, Prodiamesinae and Tanypodinae. Tanytarsini were dominant, followed by Chironomini, Tanypodinae and lastly Orthocladiinae, with very low abundance. *Tanytarsus* spp. were the most numerous and frequent taxa, while *Cladotanytarsus* representatives were the most abundant in two, high-altitude, silicate-bedrock reservoirs. Among Chironomini, the most common and abundant were *Polypedilum sordens*, *Dicrotendipes nervosus*, *Microtendipes* gr. *pedellus/chloris*, P. *nubeculosum* and *Endochironomus albipennis*. Water level fluctuations, sediment characteristics and conductivity had the highest influence on the qualitative and quantitative structure of chironomid assemblages. Most of the studied reservoirs are not under the greater influence of various kinds of pollution from land use. These results enrich our knowledge of reservoirs as aquatic habitats harboring specific fauna, including chironomids, as well as chironomid ecology and their response to environmental stress. Gathered data can be applied in creating a monitoring system for heavily modified and artificial lakes according to the requirements of the European Water Framework Directive.

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Evaluation of metal levels in soft and hard tissues of brown trout and fish intestinal parasites as indicators of wastewater impact in the karst Krka River

Assessing heavy metal pollution of rivers usually involves the evaluation of metal levels in river water and organisms as integrative long-term indicators, mostly using their liver, gills or kidney as indicator organs. Here we present a novel approach, which involves metal measurement in: a) the cell cytosol, as metabolically available fraction; b) fish calcified structures (otoliths and scales), which reflect metal exposure over a long-term period; c) fish intestinal parasites, acanthocephalans, as organisms with high metal accumulation potential and rapid response to metal changes. To assess the potential pollution of the karst Krka River by industrial and municipal wastewaters, concentrations of Fe, Mg, Mn, Rb, Sr, Tl and Zn in soft (muscle tissue, liver cytosol) and hard tissues (otoliths, scales) of brown trout (*Salmo trutta* Linnaeus, 1758), acanthocephalans (*Dentitruncus truttae* Sinzar, 1955) and water samples from a reference (river source) and a potentially polluted location were measured by HR ICP-MS (soft tissues and water samples) and LA ICP-MS (scales and otoliths). Concentrations of most metals, especially of Fe, Mn, Sr and Zn, were higher in water samples, acanthocephalans and all types of fish tissues from the polluted compared to the reference site, reflecting short- and long-term metal exposure. Exceptions were Rb and Tl concentrations, which were higher in organisms but not in the respective water samples from the reference site, so this requires further investigations. Corresponding metal levels in soft and hard fish tissues and acanthocephalans at the polluted site indicated the wastewater impact and the need of strict monitoring.

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Symbiotic bacteria from crayfish cuticle: A possibility for biocontrol of crayfish plague disease?

Microorganism *Aphanomyces astaci* (Schikora, 1906) causes the lethal disease crayfish plague, one of the reasons for decline of native decapod crayfish populationsin Europe. While *A. astaci* zoospores penetrate through the crayfish cuticle during the infection process, they encounter epibiontic bacterial communities on the cuticle surface. We have hypothesized that bacterial isolates originating from crayfish cuticle could inhibit the growth of *A. astaci* mycelium. To test this, we took cuticle swabs from five narrow-clawed crayfish, *Astacus leptodactylus* Eschscholtz, 1823 and five signal crayfish, *Pacifastacus leniusculus* (Dana, 1852) and inoculated them on PG1 medium. Next, we have isolated morphologically different individual colonies and taxonomically identified each isolate by MALDI–TOF mass spectrometry and 16S rRNA gene sequencing. Dominant bacterial genera among isolates were *Pseudomonas* and *Acinetobacter*. Furthermore, we tested the potential of isolates to inhibit the growth of *A. astaci* mycelium by plate inhibition assay. Out of 33 isolates from narrow-clayed crayfish, 20exhibited great potential for *A. astaci* inhibition, while out of 25 isolates from signal crayfish, only 2 showed great inhibition potential. This result was surprising since narrow-clawed crayfish is known to be more sensitive to *A. astaci* than signal crayfish, but could be explained by a small sample size. Further, most of the potent inhibitors from both crayfish species belonged to genus *Pseudomonas*, for which the inhibition of fish pathogenic oomycete from genus *Saprolegnia* was previously shown. In conclusion, we have characterized cultivable symbiotic bacterial communities of crayfish and shown their potential for biocontrol of crayfish plague.

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Biodiversity of Lake Crveno jezero (Red Lake) aquatic ecosystem

Lake Crveno jezero (Red Lake), located near Imotski town, Split-Dalmatia County, is a complex karst phenomenon, based on partly flooded pit, with vertical difference of -527 m, water depth ranging from 232 to 315 m and deepest point -4 m.b.s.l. It is protected in 1964 as a Geomorphological Monument of Nature, and today a Natura 2000 site (HR2000934). In 2017 and 2018, intensive ecological research was performed through two expeditions, involving speleologists, speleo-divers, biologists and geologists. Research methodology included obligate cable car installation for transport, while biological methods were adjusted for deep-water habitat. Beside microclimate measurements and photo-video documentation, we took benthic and planktonic samples, from surface until bottom. Surface water temperature varied seasonally until -20 m depth, and after, until bottom was constant (10 °C). Below -50 m depth complete darkness occurs. Riparian habitat inhabits dominantly aquatic and in small extent terrestrial taxa: Protista, Tricladida, Nematoda, Gastropoda, Hirudinea, Rotifera, Cladocera, Ostracoda, Isopoda, Hydrachnidia, Araneae, Pseudoscorpiones, Chilopoda, Diplopoda, Collembola, Trichoptera, Ephemeroptera, Coleoptera, Anura. Abundant population of endemic Spotted Minnow and Illyrian Spined Loach, both Natura 2000 taxa, occurred at all depths. On submerged walls, sessile populations of moss (*Fontinalis* sp.) and sponges (*Ephydatia fluviatilis* (Linnaeus, 1759), *Spongilla lacustris* (Linnaeus, 1759)) were recorded. In water column representatives of planktonic organisms (Protista, Rotatoria, Cladocera, Copepoda, Ostracoda) were collected, including taxa new to science, including recently described algae *Gomphosphenia plenkoviciae*. At the bottom, stygobiotic organisms (Oligochaeta, Copepoda, Amphipoda) were recorded for the first time, indicating complexity and uniqueness of this ecosystem.

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The role of dragonflies as the peak predators and indicators of the health of the Mediterranean ponds

Mediterranean freshwater ponds are specific, sometimes man-made freshwater habitats on islands and coast. Since they are mainly isolated freshwater habitats in the arid and marine environment, they represent precious habitats with high freshwater species diversity. They are sensitive to weather and detrimental human effects due to their properties (e.g. small volume, low depth, karst base, proximity to the sea). These ponds undergo washing of nutrients, toxins and soil from surrounding areas, salinization, and water level fluctuations and are often semi-permanent or temporary. Benthic macroinvertebrates are sentinel indicators of water ecological state as well as changes therein. Thus, across the Dugi otok island macroinvertebrates in 10 ponds were sampled. Benthic community was comprised of 41 taxa of which the top predators, dragonflies, were dominant, in biomass and in abundance. Therefore, further research was focused on dragonflies, specifically genera *Sympetrum* and *Anax*. Their presence differed among ponds, depending on the macrophyte coverage, salinity and the dragonfly prey size in the pond. *Sympetrum fonscolombei* was the most abundant species, with the highest biomass and thus it was the best choice for life cycle study. Head capsule width was measured with a digital micrometer and thirteen instars were distinguished and separated. Taking into account the broad taxonomic structure of the benthic community, its trophic composition, and standing stock biomass of the top predator these habitats (although in a rather high trophic state) indicated healthy and stabile ecosystems. Hence, we encourage further protection and research of these curious and precious habitats.

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New evidence supporting the role of crenal morphology in shaping water mite (Acari: Hydrachnidia) communities in karst springs

Water mites have complex life cycles, consisting of five very different life stages relating to different environmental necessity demands. Many studies have emphasized the role that water mites play within the invertebrate communities of spring habitats, both regarding species diversity and the significance within the crenal food web. In undisturbed natural springs, water mites are nearly always present and display high diversity. This study aimed to determine whether significant differences in water mite assemblages between rheocrene and limnocrene karst springs could be detected in terms of species richness, diversity and abundance, but also in different ratios of specific crenobiont and crenophilous water mite taxa. Four limnocrene and four rheocrene karst springs in the Dinaric karst region of Croatia were investigated. Seasonal samples (20 sub-samples per sampling) were taken at each spring during 2014 with a 200 µm mesh net, taking all microhabitat types with coverage of at least 5% into consideration. Water mite abundance was found to be of similar values in both spring types. Grater values of species richness and diversity indices, usually associated with rheocrenes, were indeed significantly higher in rheocrenes when compared to limnocrenes. On the other hand, unexpectedly higher shares of crenophilous and crenobiont water mite individuals (again usually associated with rheocrenes) were in this case found in limnocrenes. This unexpected discrepancy is most probably due to statistically significant differences in environmental parameters such as higher values of oxygen concentration and discharge found in limnocrenes with whom crenophilous and crenobiont taxa usually associate.

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Periphyton on different substrates in tufa depositing system

Tufa barriers have a variety of substrata, thus creating heterogeneous habitats for periphyton. We investigated the effect of three substrate types on periphyton diversity at Skradinski buk tufa barrier (Krka National Park). Samples of periphyton were taken from tufa substrate and leaf litter within experimental reach of the barrier experiencing modifications in hydrology due to the invasive species (*Ailanthus altissima* (Mill.) Swinge) removal. The dominant leaf litter species belonged to hornbeam (*Ostrya carpinifolia*). Periphyton was also sampled using artificial substrate (i.e., glass slides), placed on the same barrier, but within reach having permanent flow. Highest quantities of calcium carbonate precipitates were found on tufa substrates, while the calcite content on leaf litter and artificial substrates was lower. At tufa and leaf litter substrates periphyton sub-sampling was done by resuspension, while artificial substrate allowed direct examination. Determinations were done on live material, and showed ciliate dominance. The periphyton on the leaf litter comprised 49 taxa, while tufa substrate supported 22 taxa. Artificial substrate sustained different and the most diverse community, containing 64 taxa. Cluster analysis showed a clear separation of communities between the three substrates. Vagile periphytic taxa dominated on tufa substrate and leaf litter, while sessile taxa of suctorians and peritrichs had highest share in artificial substrate periphyton. Sampling technique can partly explain this difference, as artificial substrate analysis is the least destructive. Obtained results show high heterogeneity of periphyton within the same tufa barrier. Further research should address the periphyton diversity on artificial substrate within experimental reach.

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Long-term variation in stream physico-chemical attributes within a low-order urban stream (Veliki potok, Zagreb, Croatia)

The interplay of environmental changes brought by the various climate change and urbanisation scenarios can severely alter physical, chemical and biological attributes of streams. Intense anthropogenic activities and/or high discharge events in urban zones often result in higher in-stream loads of phosphorous and nitrogen compounds. We gathered and compared a stream physico-chemical data from three independent annual studies conducted during 1992, 1998/1999 and 2016 within a low-order urban stream (Veliki potok) located in the Medvednica Nature Park (Zagreb, Croatia). The data were collected monthly during the three study periods. The following physico-chemical parameters were compared: water discharge (obtained from Croatian Meteorological and Hydrological Service), water temperature and oxygen concentration, COD (chemical oxygen demand, i.e., a proxy for dissolved organic matter in water), and in-stream concentrations of nitrites, nitrates and orhophosphates. The data revealed *a pronounced seasonality* of water temperature and oxygen concentration with maximal temperatures in summer and minimum in winter, and the opposite oxygen seasonal trend. Significant inter-annual differences were observed only for in-stream COD and nitrate concentrations. In 1998/1999, COD levels were in average up to twofold higher than in 1992 and 2016, whereas 2016 was observed as a year of peaking nitrate concentrations, which were in average eightfold higher than in 1992 and 1998/1999. None of the water chemistry parameters was found to be correlated to the water discharge. It might *suggest an increasing anthropogenic* influence (beyond the natural variability) on the long-term variations of the investigated stream attributes within the Medvednica Nature Park.

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Reflection of hydromorphological features on the macroinvertebrate-based bioassessment of inland waters

It is not even debatable that macroinvertebrates are a component of great importance in the freshwater ecosystem. They not only play an inevitable role in energy flow through benthic food webs, but also present an excellent indicator of water quality and ecosystem health. Macroinvertebrate's indication importance is based on well investigated differences in physicochemical parameters sensitivity of each present taxon. Furthermore, macroinvertebrates were thoroughly studied and their traits are well known. They are rather large which facilitates collection and identification; they have limited areal range and a quite long life cycle suitable for monitoring environmental changes. However, scarce are the reports that assess macroinvertebrate community composition in respect to the hydromorphological traits. Is it because such correlation is not significant enough or even does not exist? Well, the aim of our study was to examine this. The area of research comprised 21 sites in Dinaric coastal ecoregion divided in 5 groups, based on altitude position, flow stability and size of waterbody. We took samples with the Surber sampler during spring and identified the macroinvertebrates to the lowermost taxonomical level. We used Asterics software for calculating bioassessment metrics and evaluation of water quality to compare them and potentially link differences within hydrological groups. Perennial and/or montainous water bodies were of higher water quality, i.e. lower saprobity, comparing to the lowland larger rivers. Accordingly, the former habitats support higher diversity and number of taxa.

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The effects of three psychoactive compounds on behavior of the invasive signal crayfish, *Pacifastacus leniusculus* (Dana, 1852)

Invasive species and pollution are one of the dominant constituents of anthropogenic environmental disturbance with major economic and ecological impacts. However, the interactive effects of myrad of chemicals in aquatic environments on invasion rates and success are currently poorly understood. The aim of this study was to examine whether psychoactive compounds that are considered as contaminants of emerging concern and are ubiquitous in the Sava River (2 antidepressants and one illicit drug) could influence the behavior of the most successful invader of European freshwaters - the signal crayfish (*Pacifastacus leniusculus*). Through a week-long trial, we exposed crayfish to i) environmental concentrations and ii) therapeutic concentrations of citalopram, nortryptiline and amphetamine and recorded their behaviors after 24 hours and a week-long exposure. We recorded specific parameters (velocity, distance moved, moving and non-moving cumulative duration) using EthoVision programme and compared the behavior of exposed crayfish to their respective size matched controls. Out of three analysed compounds, citalopram and amphetamine had a significant effect on crayfish activity but only after 7 day exposure. Overall, citalopram decreased while amphetamine increased cumulative duration of movement. We discuss our finding in the context of their potential effects on physiology and fitness of signal crayfish.

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New species of Calanoid copepod of genus *Eudiaptomus* Kiefer 1932

The Western Balkans is recognized as a European biodiversity hotspot, with high endemism particularly in subterranean and freshwater fauna, with coldwater springs and ancient lakes identified as regional hotspots of freshwater diversity. In microcrustaceans, awareness of the diversity of groundwater fauna has increased in recent decades. However, the region is still understudied, and many cryptic species have been recently discovered in different freshwater taxa. Copepods are abundant and diverse in fresh waters. The Diaptomidae is the largest family within the order Calanoida. Many genera of Diaptomidae are endemic to particular continental areas, like the genus *Eudiaptomus*, having the highest species richness in the Palaearctic region. There are 29 *Eudiaptomus* valid species. In Europe, there are two endemics, *Eudiaptomus padanus* (Burckhardt, 1900) and *Eudiaptmus hadzici* (Brehm, 1939), with ranges restricted to potential glacial refugia, the Apennine and the Balkan Peninsula, respectively. In this study three new species of Calanoid copepod of genus *Eudiaptomus* Kiefer 1932 are described, and redescription of *Eudiaptomus hadzici* is given. The genus now includes three new species, all endemic to south-east Europe. The male of four species can be distinguished by morphology of the exopodite and endopodite of leg 5, and spine on segment 13 of right antennule geniculate. Key to the four species of genus *Eudiaptomus* is provided.

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Negative effects of introducing allochthonous species *Pacifastacus leniusculus* (Dana, 1852) into aquatic ecosystems of Bosnia and Herzegovina

The problem of introducing allochthonous species into watercourses has already had negative impacts on ecosystem services and their benefits. The most vulnerable parts of watercourses are those that are already under the anthropogenic influence (aquaculture, hydropowers, waste waters, etc.). Expansion of allochthonous species (often invasive) is most often the result of intentional input due to species attractiveness and insufficient awareness of the risks of such activities. Allochthonous species *Pacifastacus leniusculus* (Dana, 1852) was recorded in the Una River in October 2018, as a first decapod allochthonous species in Bosnia and Herzegovina freshwaters. According to the longitude characteristics, the species has been recently introduced and has failed to increase the population. The river Una is a habitat of white-clawed crayfish, and in the tributaries Sana, Sanica, Biljanska river, Banjica and the streams near Ribnik, a large population of *Austropotamobius torrentium* (Schrank, 1803) were ascertained. In the river mouth of Una River, the introduction of native species of *Astacus leptodactylus* Eschscholtz, 1823 was carried out. Negative effects primarily affect the food chain by increasing consumer rates (predators). There is a significant impact on macrophytes communities, the zoobenthos fauna, crayfish plague appearance, and the possibility of spreading this very invasive species to the other parts of the river Una and its tributaries, which will have a direct effect on the indigenous species of decapods. There is no state institution for the monitoring and control of invasive species, which is a major problem in defining protection measures and adequate management of watercourses.

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Changes in the periphytic ciliate community structure under different hydrological conditions in a temperate floodplain lake

Ciliates represent important component of periphytic communities in aquatic habitats, including river-floodplain ecosystems, particularly as a link between lower and higher trophic levels in food webs. Considering that these assemblages have been poorly studied in floodplain habitats, the aims of this research, conducted in a Danubian floodplain lake (Kopački Rit Nature Park, Croatia), were to identify the main factors controlling periphytic ciliates and to estimate their ecological role in this dynamic ecosystem. The study included two experimental series lasting from spring until winter and from summer until winter in 2010. The glass slides were chosen as artificial substrates for periphyton development. Floods of different intensity and duration characterised the research period and modified the lake water properties. During the flooding, water transparency and nutrient concentrations increased, while conductivity and chlorophyll *a* concentration decreased. Macrophyte and metaphyton stands spread out within the lake during the high floods and periphyton biomass decreased consequently. In such conditions, the total ciliate abundance in periphyton declined. Peritrichs, sessile filter feeders that utilize suspended prey from the surrounding water, were dominant ciliates in the periphyton, with the highest densities registered in the absence of floods. However, during the extremely high water-levels, the abundance of mobile raptorial and filter-feeding ciliates increased. Bacteria and algae represented the main food source for periphytic ciliates. This study reveals the importance of hydrological regime in shaping periphytic ciliate community and suggests that these organisms have a significant impact on plankton communities in temperate floodplain lakes.

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Indirect influence of pesticides on macroinvertebrate community of constructed lakes

Synthetic pesticides are substances used to control pests of both animal and plant origins on agricultural land. These substances often find their way to aquatic ecosystems by runoff where their effect on non-targeted aquatic invertebrates, plants and fish can range from individual to community level. In constructed lakes created by damming, the risk of accumulating these harmful substances is potentially greater than in other surface waters. Pesticide runoffs occur directly in the drainage basin of the artificial water bodies and along the longitudinal drainage surface of the tributary running water(s). Our main hypothesis was that persistent exposure even to "acceptable pesticide levels" alter the aquatic macroinvertebrate community equilibrium, decreasing the abundance of specific predator groups with longer generation time (namely dragonflies and water mites), favoring thus r-type selected species. Preliminary screening samples of macroinvertebrate communities were taken in autumn of 2016 from 10 constructed lakes located in the agricultural-land-dominated Pannonian Lowland Ecoregion of Croatia. Ten subsamples at each site were collected using benthos hand net (500 µm). Monthly measurements on eight pesticide types were collected from 2010-2016 in tributary rivers close to the mouth of the constructed lakes. Significant correlations were determined between the share of agricultural land cover present in the drainage area and pesticide concentrations. A multiple linear regression model showed that r-dominance (ecological quality bioassessment metric) increased with increasing pesticide concentrations. Increased concentration of pesticides was also found to be detrimental to predator groups with longer generation time and a smaller number of offspring (K-selected species).

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Re-discovering the aquatic flora of Neretva River Delta

Neretva River Delta is a marshland area situated in southern Dalmatia, encompassing the last 20 km of the river, together with a network of surface water bodies, which are well interconnected due to the long-term human influence and interventions into the landscape. Previous studies revealed over 800 plant taxa in the Delta, however no systematic study of waterbodies was ever performed. In this study, we have focused on mapping two alien aquatic plants *Egeria densa* and *Myriophyllum heterophyllum* along the waterbodies in the Delta, by performing a systematic and comprehensive fieldwork. We have recorded a large number of new localities of these aliens and observed populations displaying invasive behaviour. Apart from the targeted species, we have recoded 62 mostly aquatic plants. Altogether eight threatened taxa were recorded: critically endangered (CR) *Dorycnium rectum* and *Hydrocotyle vulgaris*, endangered (EN) *Cynanchum acutum*, *Hippuris vulgaris*, *Hottonia palustris*, *Periploca graeca* and *Ranunculus lingua* and vulnerable (VU) *Cyperus longus*. Near threatened (NT) *Butomus umbellatus* and data deficient (DD) *Utricularia australis* were also recorded. The species *Hydrocharis morsus-ranae* and *Nymphoides peltata*, not previously known from the Mediterranean Croatia apart from one and two historical records from Neretva, were found on many sites during this study. Similarly, we have recorded *Sagittaria sagittifolia* and not previously known from Mediterranean Croatia and *Hottonia palustris* and *Thelypteris palustris* previously recorded in Dalmatia only once. A rare species *Cladium mariscus* was also recorded, as well as a very rare moss *Riccia fluitans*, previously known only from very few localities in continental Croatia.

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Diversity of the aquatic plants in the *Herbarium Croaticum* (ZA) collection (Zagreb, Croatia)

The aim of this study was to collect and analyse data on aquatic plants within the *Herbarium Croaticum* (ZA), the largest and oldest herbarium collection in Croatia. In total 951 herbarium sheets representing 72 aquatic taxa were renovated, digitized, geo-referenced (generating distribution map for each taxa) and made publically available in *Flora Croatica Database* and *Virtual herbarium of ZA & ZAHO*. Aquatic plants within ZA collection have been collected during the period of 165 years, the oldest sheet originates from as early as 1844, while the average age of herbarium sheets is 76 years. The most abundant genera are *Potamogeton* and *Ranunculus*, whereas most common families are Potamogetonaceae and Ranunculaceae. Most of the specimens were collected in Croatia, and smaller part in neighbouring and geographically close countries (Slovenia, Hungary, Serbia, Bosnia & Herzegovina, Montenegro, Italy, Austria and FYR Macedonia). As much as 28 Red listed aquatic taxa are stored within ZA collection. The importance of collection is expressed through specimens of many rare and threatened species, for which this collection represents the only evidence of their presence in Croatia (regionally extinct *Caldessia parnassifolia*, as well as *Luronium natans*, *Callitriche platycarpa*, *C. truncata*, *C. hermaphroditica*, *Potamogeton alpinus*, *P. polygonifolius*, *Scheuchzeria palustris*, *Rynchospora fusca*, *Myriophyllum alterniflorum*, *Nuphar × spenneriana* and *Sparganium minimum*). Collection of aquatic plants in ZA is important source of data considering their historical and recent distribution, which are the basis for estimation of distribution changes, threat evaluation and protection planning.

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Microphyte community structure on *Ceratophyllum demersum* in drainage channels (Baranja, Eastern Croatia)

Macrophytic vegetation present in various aquatic ecosystems provide a good substrate for the development of epiphytic algae and cyanobacteria. A macrophyte *Ceratophyllum demersum* is particularly well developed and has great coverage in channels characterised by low water velocity. The research on microphyte diversity and abundance on *C. demersum* was conducted in three drainage channels in Baranja (Eastern Croatia) in September 2016. Different values of physico-chemical water properties were found in investigated biotopes. Higher concentrations of oxygen (9.72 mg/L), total phosphorus (0.88 mg/L) and higher conductivity were found in the shallowest channel in Jagodnjak, while two other channels (near Eblin and Darda) had a higher concentration of nitrogen compounds. The highest content of chlorophyll *a* in water (41.61 µg/L) and epiphyton (Chl-a: 509.19 ± 67.88 µg/gDW), as well as high microphyte diversity (94) and abundance (555 672,05 x 10^3 ± 49 261,11 x 10^3 cell/gDW) was found in the channel near Eblin. Cyanobacteria *Heteroleibleinia ucrainica* was the most abundant at all localities accompanied by *Tolypothrix* sp. in the channel near Darda. An invasive and potentially toxic cyanobacteria *Cylindrospermopsis raciborskii* was found near Eblin. The results indicate that despite differences in the physico-chemical environment and channel hydro-morphology, the substrate type is one of the most important factors conditioning epiphyton development and structure in eutrophic aquatic biotopes.

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Benthic invertebrates, fish and zooplankton coupling in freshwater Mediterranean ponds

Mediterranean coastal and islands region abounds in small and shallow, mainly temporary freshwater ponds which have been rare subject of a scientific topic for a long time. They represent unique habitats in dry and arid climate inhabited numerous species, especially invertebrates with a short life cycle and can be rapidly colonized during the flooding season. Impact of benthic invertebrate predators and fish on zooplankton was assessed in the island freshwater ponds (island Korčula), and this is one of the first data for these habitats in the eastern Adriatic coast. The goals of this study were to investigate: i) interaction among benthic invertebrates, fish and zooplankton; and ii) difference in shelter selection, bottom sediment *vs.* macrophytes. Analysis of ecosystem functioning shows decreased abundance of planktonic crustaceansin ponds with low macrophyte coverage, and *vice versa*, which highlighted role of macrophyte for zooplankton refuge against predators (acari, heteropterans, fish). Results suggest that macrophyte can serve as shelter for zooplankton, but in the case of high macrophyte abundance and numerous benthic invertebrate predators, role of macrophyte shelter is diminish, and zooplankton probably found refuge in sediment. This is confirmed by increased abundance, especially cladocerans, in the sediment. Through the dual impact of predators and low macrophyte coverage, as well as huge impact of benthic invertebrates among macrophytes, sediment was an efficient refuge for zooplankton. Interaction between predators (benthic invertebrates and fish) and zooplankton displayed higher complexity along habitat heterogeneity, while zooplankton share an important fraction in food resources among the trophic network.

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The effects of psyhoactive substance mixture on behaviour and physiology of invasive signal crayfish, *Pacifastacus leniusculus* (Dana, 1852)

It is known that the presence of psychoactive substances, such as antidepressants and drugs is relatively constant in freshwater ecosystems of urbanized areas. Recent studies have demonstrated their bioaccumulation in the tissues of aquatic organisms and effects on their behaviour and physiology in environmentally relevant concentrations. However, their potential interactive effect, especially on invertebrates, is not sufficiently explored. This study aims to assess whether the mixture of two antidepressants and an illicit drug commonly found in the Sava River influences the behaviour and neurophysiology of the most successful crayfish invader in Europe - the signal crayfish (*Pacifastacus leniusculus*). Within a weeklong trial, we exposed crayfish to environmental concentrations of citalopram, nortriptyline and amphetamine dissolved in ambient water. We measured behavioural and physiological endpoints: i) the effects of 24h and 7 day exposure on dynamics of antagonistic interactions between size-matched pairs of exposed and non-exposed (control) crayfish, ii) the effects on serotonin levels, iii) the effects on glucose levels in haemolymph and iv) biomarkers of oxidative stress. We did not observe any statistically significant difference in the crayfish behaviour except that exposed individuals in average took slightly longer time to engage in fights. We observed a significant increase of lipid peroxidation in exposed individuals, suggesting potentially an adverse synergistic impact of examined pharmaceuticals on crayfish, in environmentally relevant concentrations.

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Aquatic vegetation of Lake Visovac

Lake Visovac is situated in Krka National Park and it represents the largest extension of Krka River. In summer 2018 the line transect sampling method was applied for vegetation mapping. In total 21 transcts perpendicular to the shore up to the depth at which the macrophytes cease to appear were made, generating as much as 293 species/location records. In total, 26 species of aquatic plants was found, including one yellow-green algae, eight charophytes, two mosses and 15 vascular plants. The most common species in the flora of Lake Visovac are *Myriophyllum verticillatum*, *Najas marina*, *Phragmites australis* and *Potamogeton perfoliatus*. Discovery of a rare aquatic moss *Fissidens fontanus* is of great importance, being the second in Croatia. There are four main vegetation belts in the aquatic vegetation of Lake Visovac: helophytic (dominantly reed) vegetation, submerged vegetation with or without floating leaves (belonging to NATURA 2000 habitat type 3150 Natural eutrophic lakes with Magnopotamion or Hydrocharition) and charophyte vegetation (NATURA 2000 habitat type 3140 Hard oligo-mesotrophic waters with benthic vegetation of *Chara* spp.). Lake Visovac is dominated by plants of light, warm habitats which survive unfavorable living conditions with their buds in water and which prefer supstrata rich in nutrients and water with neutral pH and low salinity. The increased inlet of nutrient rich water and the anchoring represent the main threats to the preservation of Lake Visovac vegetation.

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What are key factors defining phytoplankton communities in two connected karstic Mediterranean lakes?

The Baćina lakes represent a complex of six connected and one isolated lake. Samples for physical, chemical and phytoplankton analysis were collected as composite samples from April until September 2017 in two deepest and largest lakes, Crniševo and Oćuša, with the aim of defining taxonomic and functional differences in phytoplankton composition. Temperature and oxygen profiles were measured every meter. In the Lake Crniševo, stratification was present throughout the whole study period while in Lake Oćuša stratification was not detected during spring. Bottom hypoxia was recorded in April and May in the Lake Oćuša while during the other months anoxia was recorded in both lakes. In springtime (prior stratification), representatives of the functional group B sensitive to this phenomenon were dominant in the Lake Oćuša, unlike codominance of functional groups X2, L0 and F, tolerant to the stratification present through all investigation period in the Lake Crniševo. Statistical analysis indicates that differences in phytoplankton communities in lakes Oćuša (B → L0, F → X2) and Crniševo (X2, L0 → F → F, L0) are not defined with general water physical and chemical properties but with nutrient deficiency and stratification. Longer period of investigation is suggested with the aim to understand better the processes in these lakes.

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Preliminary results of research in "Tomina jama" anchialine cave, Neretva River Estuary, Croatia

A preliminary research of Tomina jama cave was conducted in 2016 and 2017.Tomina jama is currently the only known anchialine cave in the Neretva estuary. The cave has been explored up to 58 m with the last 25 m submerged. Tomina jama is a typical anchialine cave with a freshwater lens at the surface and seawater layer at the bottom. Salinity in the cave increased from 0 at the surface to 33 ‰ at a depth of 22 m as dissolved oxygen decreased from 9.1 mg L^{-1} to anoxic conditions at depth. The hypoxic/anoxic zone extended for more than 10 m. Water temperature ranged from 14.8 °C at the surface to 16.2 °C at the bottom while pH decreased with depth from 7.74 to 6.86. Several stygobionts were discovered in the freshwater layer including Serpulids *Marifugia* sp. and the crustaceans *Monolistra* sp., *Troglocaris* sp. and *Niphargus* sp. Prior to this discovery, *Marifugia* have only been found in freshwater caves from Dinaric karst region. Preliminary DNA results indicate that this species is not *Marifugia cavatica* Absolon & Hrabe, 1930 and may be a newly discovered species. Below the halocline, the cave walls and bottom were lined with a significant bacterial mat. This cave system is the furthermost inland of any Croatian anchialine cave with significant communities in both the fresh and saltwater layers. The discovery of *Marifugia* sp. populations and the bacteria mats require further research.

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Sustainability of freshwater salmonid farming in a karst river ecosystem

Eutrophication in freshwater ecosystems is one of the major environmental problems. Aquaculture industry is greatly impacting aquatic environments by introducing nutrients and microorganisms through nutrient enrichment of water column and sediment. The aim of this research was to evaluate the impact of a salmonid farm on the environment. Water and sediment were sampled once a season from spring 2016 to spring 2018 at the commercial salmonid farm in Šibenik-Knin County. Samples were processed for bacterial indicators (total coliforms, *Escherichia coli*, enterococci and *Pseudomonas aeruginosa*, total number of bacteria). Physicochemical parameters of water on the salmonid farm and the surrounding environment were analysed, as well as granulometric and mineral composition, carbonates and organic matter in the sediment. The objective was to determine water quality above the fish farm, in the fish farm and below the discharge of water in the river, and sediment characteristics in the fish farm and downstream of the fish farm. Water and sediment quality indicators were statistically analysed by Principal component analysis (PCA) in order to determine any grouping regarding locations and seasons. Results for the water do not differentiate fish farm location from the other sampling points, while sediment results show a mild trend of separating fish farm regarding total coliforms and total number of bacteria. Sedimentology analysis detected somewhat higher organic matter content in the farm sediment (4.05-4.18%), indicating its increased intake compared to the river (1-1.5%). This indicates that the impact of salmonid aquaculture is limited in the farm boundaries.

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Effects of UVB-Radiation and Norflurazon on Mesoglea in Green Hydra (*Hydra viridissima* Pallas, 1766) and in Brown Hydra (*Hydra oligactis* Pallas, 1766)

Symbiotic green hydra (*Hydra viridissima* Pallas, 1766) and non-symbiotic brown hydra (*Hydra oligactis* Pallas, 1766) were exposed to the UVB-radiation and/or herbicide norfluorazon in order to determine their effect on extracellular layer mesoglea. The experiment was performed using crystallizing dishes 60 ml volume with either 50 ml of aquarium water or with two norflurazon solutions ($2x10^{-6}$ and $2x10^{-7}$ mol/L). Ten individuals of green and brown hydra respectively were placed in separate dishes and five individuals of each species were placed together in a dish. All specimens were exposed to UVB-rays with a wavelength of *254 nm* for two minutes. The second part of the experiment was performed in the same way except there was no UVB-radiation. Hydras placed in aquarium water were not exposed to the UVB-radiation, as a control group. The experiment was performed in triplicate for 72 hours and hydras were monitored every 24 hours. For slide preparation, standard histological methods were used and slides were stained with hematoxylin and eosin. ImageJ software was used for the morphometric measurements. Thickness of mesoglea was measured at 30 randomly selected segments on the histological slides, per particular treatment. Mesoglea was least thick in the samples of green and brown hydra exposed to UVB-radiation, while the greatest thickness was noted in the group of both species treated with norflurazon concentration of $2x10^{-7}$ mol/L. The results indicated that xenobiotics could have an impact on alteration of mesoglea thickness in hydra, which can affect physiological and regenerative processes in symbiotic and non-symbiotic hydras.

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The importance of regular museum collections processing: case of water beetles (Insecta: Coleoptera) in Karaman and Novak collections, Natural History Museum in Split

Natural history museums are both cultural and scientific institutions of indispensable importance, especially nowadays when biodiversity loss increases. The Natural History Museum in Split was founded in 1924, where, besides the other, two valuable entomological collections with approximately 35000 museum objects and 9000 taxa are held: Beetle Collection of Eduard Karaman and Entomological Collection of Petar Novak. Both collections originated at the beginning of the 20th century. Water beetles are widespread group of aquatic insects inhabiting all types of aquatic habitats, and are often used as bioindicators in water quality assessments. The aim of this study was to analyse water beetles' data in the entomological collections in following steps: i) to list the objects; ii) to revise their identification; iii) to revise the nomenclature; iv) to analyse its geographical affiliation. In total, 1100 museum objects of water beetles were listed representing 338 taxa. The most speciose families were Dytiscidae and Hydrophilidae. Five new species for Croatian fauna were found, but with uncertainty as valid identification is impossible as genitalia were not extracted during the preparation process. Beetles were collected mostly in Dalmatia, in a period from 1896 to 1917. Results indicate occurrence of high species diversity in that area, as well as the presence of nowadays potentially rare and/or endangered species. Future research should be focused on the comparison of estimated fauna in the Museum with the fauna of water beetles that inhabits same area nowadays, all with a vision to the future protection of Croatian aquatic fauna.

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Growth inhibition and recovery of aquatic macrophyte *Lemna minor* after pulsed herbicide exposures

During and after precipitations, herbicides can easily reach aquatic ecosystems via agricultural runoff, drainage and leaching processes. Consequently, the exposure of non-target aquatic organisms to herbicides occurs as multiple pulses. This type of exposure is characterized by fluctuating herbicide concentrations and pulse exposures followed by periods of recovery. Nevertheless, such exposure scenario is not considered in standardized laboratory ecotoxicity tests. The aim of the present study was to investigate the effects of isoproturon on the growth and recovery of common duckweed (*Lemna minor* L.) over a 14-day period after 48 h of pulse exposure, as well as comparison with effects observed in standard OECD *Lemna*-test with continuous exposure. Isoproturon is a selective and systemic phenylurea herbicide. It represents one of the most widely used herbicide in intensive agricultural production. Duckweed plants were treated with 0.05, 0.10 and 0.20 mg L^{-1} of isoproturon in nutrient solution. Complete recovery of plants treated in two 48 h pulses with 0.05 and 0.10 mg L^{-1} of isoproturon was achieved within 14 days. In the treatment with 0.20 mg L^{-1} of herbicide there was a slight growth recovery, but plants did not completely recover, whereas the most toxic isoproturon activity was established at all tested concentrations in continuous exposure. The results from this study suggest that recovery is possible for non-target primary producers. The potential for aquatic organisms to recover after pulsed herbicide exposures should be more actively considered in order to better predict the ecological risk assessment of agrochemicals.

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Hydra vs. turbellarians: who is the strongest constituent in a given micro-(eco)system? – preliminary observations

The goal of this research was to study interactions among carnivorous invertebrates: green hydra (*Hydra viridissima* Pallas, 1766), and turbellarians *Dugesia gonocephala* (Duges, 1830) and *Polycelis felina* (Dalyell, 1814). In *ex situ* microcosms experiment animals were exposed through the day-night rythm (8 h light/16 h dark) and different light and temperature (25 °C/13.5 °C) conditions, both separately with starved and fed animals. The results were recorded immediately after the experiment setup and every 1, 8 and 24 hs after the beginning of the experiment, including the controls. Experiment was set up with 1 and 5 animals of each species, 5 replicas for each setup. In the view of interference competition, behaviour indicated habitat separation: fed *D. gonocephala* was resting on the side and hydra on the bottom of microcosm, with partial migrations in water column. Starved *D. gonocephala* were agile and hydra was on the bottom of microcosm, with no migrations present. In microcosm with fed *P. felina*, hydra was budding and *P. felina* were mainly resting. Starved specimens of *P. felina* were active and hydras were relaxed. Interspecies predation occurred between *D. gonocephala* as a predator and *P. felina* as a prey, mainly observed with starved animals. *P. felina* used aggregation as the mechanism of protection. Results of this study suggested hydra as better-equipped predator, probably due to cnidocytes, than turbellarians. Two or more predatory groups can coexist, however closely related interspecies predation could appear.

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Ecological effects of increased water level and salinity variations in the shallow Lake Vransko near Biograd n/M

Lake Vransko, the largest natural lake in Croatia by surface area, was declared a Nature Park in 1999. This shallow lake (max depth -3.9 m a.s.l.) is located in the karst in close proximity to the Adriatic Sea to which it is directly connected by the Prosika canal, dug in the 18th century. Climate change (seawater level rise and decreased precipitation) and increased water usage in the catchment area caused oscillation of freshwater level resulting in increased lake salinity due to seawater inflow directly through Prosika canal and indirectly through the karst ridge between the lake and the sea. Water level range and salinity variations (<1-4 ‰) increased remarkably during the last two decades, especially during dry years (max. recorded salinities: 2008 – 11.3‰; 2012 – 18.2‰). At that time, also the most pronounced minimums of water level was observed in more than 50 years of data records. The aim of this work is to present all known ecological effects of water level and salinity variations in Lake Vransko. Hydrological and physicochemical conditions for the period 2000-2017 were analyzed and presented. Biological monitoring data was used to show how periods of increased salinity affected planktonic, benthic (littoral macroinvertebrates and macrophytes) and fish assemblages in the lake. A reconstruction of Prosika canal with movable gate construction is planned in order to prevent water level decrease and seawater inflow during dry years. Different groups of organisms can be used as indicators for an adaptive water management plan implementation after the reconstruction.

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Ludwigia peploides (Kunth.) P.H. Raven - A new species in Croatian Flora from the list of invasive alien species of Union concern

The species *Ludwigia peploides* – the floating water primrose, is a perennial macrophyte. It is naturally distributed in South and Central America and the southern part of the USA. According to some databases it is also naturally distributed in Australia and New Zealand. The floating water primrose was imported in Europe during 1830's in Montpellier (France), probably as an ornamental species which spread and became invasive. Since then it was recorded in the following European countries: Belgium, France (Corsica), Greece, Italy, Netherlands, Spain, Switzerland, Turkey and the UK where its distribution is limited or its invasiveness is at an early stage. *Ludwigia peploides* is one of the species listed as IAS of EU concern (Regulation EU No 1143/2014) and a new species for the Croatian flora. It was recorded at the end of August 2018 on the Ilova River near the village of Kaniška Iva (Moslavina, Croatia) where it forms dense floating mats. Its presence was observed along a length of the river of about 2 km. The way of its entrance to the territory of Croatia is not known. However, a possible pathway may include the availability of internet purchase of floating water primrose for aquarium or a garden pond purposes from which it could have spread into natural habitats. With this finding there are now six plant species listed as invasive alien species of Union concern present in Croatia.

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Review of water dynamics in Europe due to climate change

Natural river flow regimes in Europe have already been heavily modified by various anthropogenic impacts such as damming, channelization and water withdrawals. Climate change is an additional factor, which is likely to have a stronger impact on flow regime modification than dam operation and water abstractions have had up to now. The main climate elements whose changes will affect the hydrological cycle and river flow regimes are precipitation redistribution (both by geography and by intensity), temperature increase, and snow cover melt. The changes of these elements and their consequences will exhibit different patterns across Europe. The highest degree of change can be expected in the Mediterranean and Boreal climate zone. It is observed that a changing climate has shifted the timing of river floods in Europe during the past 50 years. In northeastern and western Europe there is a shift toward earlier floods. However, around the North Sea and in some parts of the Mediterranean coast, timing has shifted toward later floods. The temperate oceanic zone will probably be least affected, in response to the more moderate changes in climate variables. Although, in the Netherlands, some effects such as rising sea level and increase in river discharge are already obvious and may cause serious problems in the future. Climate change will have major consequences both on land and the ecosystems and on the availability of fresh water for human consumption, so it is imperative to carefully manage water resources in order to alleviate them.

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Predation in turbellarians: *Dugesia gonocephala* as superior predator - preliminary observations

Turbellarians (flatworms, Platyhelminthes) in freshwater or moist terrestrial environments are mainly predatory species. In these experimental setups, we assessed interaction between turbellarians *Dugesia gonocephala* (Duges, 1830) and *Polycelis felina* (Dalyell, 1814) in terms of their competitive capability. In *ex situ* microcosms experiment turbellarians were exposed through the day-night rhythm to different temperature and light conditions: 25 °C and 8 h light, 13.5 °C and 16 h dark, respectively. The results were recorded immediately and every 1, 8 and 24 h after the beginning of the experiment. Five replicas with both fed and starved animals were set up. Species *D. gonocephala*, in both conditions of temperature and light, attacked and preyed upon *P. felina* and took over pigmentation from *P. felina* considering that *P. felina* is black colored and *D. gonocephala* is grey transparent. There was no reverse process recorded. Moreover, it was noticed that previously fed *D. gonocephala* specimens more likely attacked *P. felina* than starved individuals did. Also, the higher rate of predation was present at higher temperature (25 °C) than under the conditions of lower temperature (13.5 °C). Generally, when turbellarians were put in microcosm at the beginning of the experiment, they were very agile searching for prey. Afterwards, *D. gonocephala* and *P. felina* rested mostly on the opposite sides of microcosms. Results of our study indicated new insights into predation among turbellarians and more information about freshwater micro-ecosystem food competition.

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The relevance of ITS region as a genetic marker in determination of *Pomphorhynchus* species (Acanthocephala) from Croatian rivers

Acanthocephala are intestinal helminths of fish, interesting as a potential sensitive biological indicator of metal bioavailability in river water. Their morphological identification is difficult because main taxonomic characters are often hardly visible in fixed specimens and show considerable intraspecific variation. The aim of our study was to check if ITS region is a reliable genetic marker for identification and differentiation of acanthocephalan members of the genus *Pomphorhynchus* from the freshwater fish in Croatian rivers. Individual parasites were collected from the different fish species in rivers which belong to the Black Sea basin (Sutla, Sava, Kupa, Dobra, Mrežnica, Korana, Orljava, Una and Danube). After morphological determination, ITS region was amplified by PCR, followed by commercial sequencing and bioinformatics analyses. Three species, *P. laevis*, *P. tereticollis* and *P. bosniacus* were identified by morphology. Two species, *P. laevis* and *P. tereticollis* were clearly differentiated by ITS region, while the third species, *P. bosniacus* had the identical or very similar ITS sequence to *P. laevis* specimens. Results of our study confirmed ITS region as a good tool for differentiation of two sister species, *P. laevis* and *P. tereticollis*. They also indicated *P. laevis* as a complex, the most abundant and widely distributed species in explored rivers within *Pomphorhynchus* genus.

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A long-term study of mayfly emergence patterns in the Dinaric Karst freshwater habitats

Mayfly emergence patterns were studied over a ten-year period in two types of karst freshwater habitats: a spring and two tufa barriers, using pyramid-type emergence traps. A total of nine mayfly species were recorded. The highest proportion of collected individuals belonged to the genus *Baetis* Leach, 1815 which was recorded at all three study sites, but we were unable to distinguish between two included species (*B. rhodani* (Pictet, 1843) and *B.* cf. *nubecularis* (Eaton, 1989)). Other numerous recorded species were *Paraleptophlebia submarginata* (Stephens, 1835)*, Ephemera danica* Müller, 1764 and *Rhithrogena braaschi* Jacob, 1974. Tufa barriers had higher species richness but lower population densities compared to the spring. NMDS analysis separated spring from tufa barriers. In the studied spring, emergence mainly occurred between March and November, and the main trigger for emergence was photoperiod. In tufa barriers, emergence mainly occurred between April and July and was related to the elevated water temperature. Generally, a higher abundance of emerging individuals was recorded during the years with higher water discharge. Emergence patterns of some species were in accordance with their typical Central European emergence patterns (e.g. *E. danica*) while some other showed certain discrepancies (e.g. *Rh. braaschi*).

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