TRANSYLVANIAN REVIEW OF SYSTEMATICAL AND ECOLOGICAL RESEARCH

19.2

The Wetlands Diversity

Editors

Doru Bănăduc & Angela Curtean-Bănăduc

Sibiu - Romania
2017

TRANSYLVANIAN REVIEW OF SYSTEMATICAL AND ECOLOGICAL RESEARCH

19.2

The Wetlands Diversity

Editors

Angela Curtean-Bănăduc & Doru Bănăduc

"*Lucian Blaga*" University of Sibiu,
Faculty of Sciences,
Department of Ecology and Environment Protection

Published based mainly on some of the scientific materials presented at the sixth
"Aquatic Biodiversity International Conference" – Sibiu/Romania 2017

ESENIAS
East and South
European network for
Invasive Alien Species

Ecotur Sibiu
N.G.O.

"*Lucian Blaga*"
University
of Sibiu

International
Association for Danube
Research

Sibiu - Romania
2017

Scientifical Reviewers

Editorial Assistants

IN MEMORIAM

Dan Munteanu
(1937 – 2017)

The Romanian biologist *Dan Munteanu* was born in Cluj, in Transylvania, on the 2nd of June 1937.

After graduating from the Natural Sciences Faculty at Babeş-Bolyai University of Cluj, in 1969 he obtained his Doctorate in Biology at the University of Bucharest by defending his doctoral thesis on the Bird fauna of the Mountain Areas of the Moldavian Bistrița River.

A complex personality, *Dan Munteanu* was known in scientific circles as one of the most competent and active ornithologists of Romania, with a solid and wide-ranging background in classical biology.

He chaired the Romanian Ornithological Society; he was member of the Executive Board of the International Waterfowl and Wetlands Research Bureau of Slimbridge (UK); and was representative of the International Council for Bird Preservation of Cambridge (UK). In this last quality he took part in a variety of European nature conservation and preservation programmes.

His bird fauna studies represent a comprehensive list of works, not only in terms of numbers but also of quality, the most important being the following: Provisional Atlas of Romanian breeding birds, a work carried out for the international committee responsible of the European Atlas; the Birds Chapter in the Romanian Red Book of Vertebrates, in the Editions of the Romanian Academy, 2005; Romanian Bird Areas – Documentations, ALMA MATER Editions, Cluj-Napoca, 2004; Rare, vulnerable and endangered birds in Romania, ALMA MATER Editions, Cluj-Napoca, 2009; Romanian Fauna, Aves, Volume XV, Fascicule 2, Editions of the Romanian Academy, 2015.

Dan Munteanu, as a member of the Romanian Academy, was also very effective in advising on broader environmental issues faced by Romania.

Starting with 2000, he chaired the Commission for the Protection of Nature Monuments of Romania, and solved very competently all problems related to its organisation and functioning. In the same quality, he coordinated the activity of the Romanian Scientific Councils of Nature Parks and National Parks. He contributed substantially to the establishment of new protected areas and proposed viable solutions for the conservation of biodiversity and the protection of natural heritage.

The death of *Dan Munteanu*, after a long and painful illness, is a great loss for Romanian biology in the many important areas to which he dedicated his tireless lifelong activity.

A generous and modest man, who dedicated his life to the protection of nature, has left us and will be greatly missed.

The Editors

CONTENTS

Preface

In a global environment in which the climate changes are observed from few decades no more only through scientific studies but also through day by day life experiences of average people which feel and understand allready the presence of the medium and long-term significant change in the "average weather" all over the world, the most comon key words which reflect the general concern are: heating, desertification, rationalisation and surviwing.

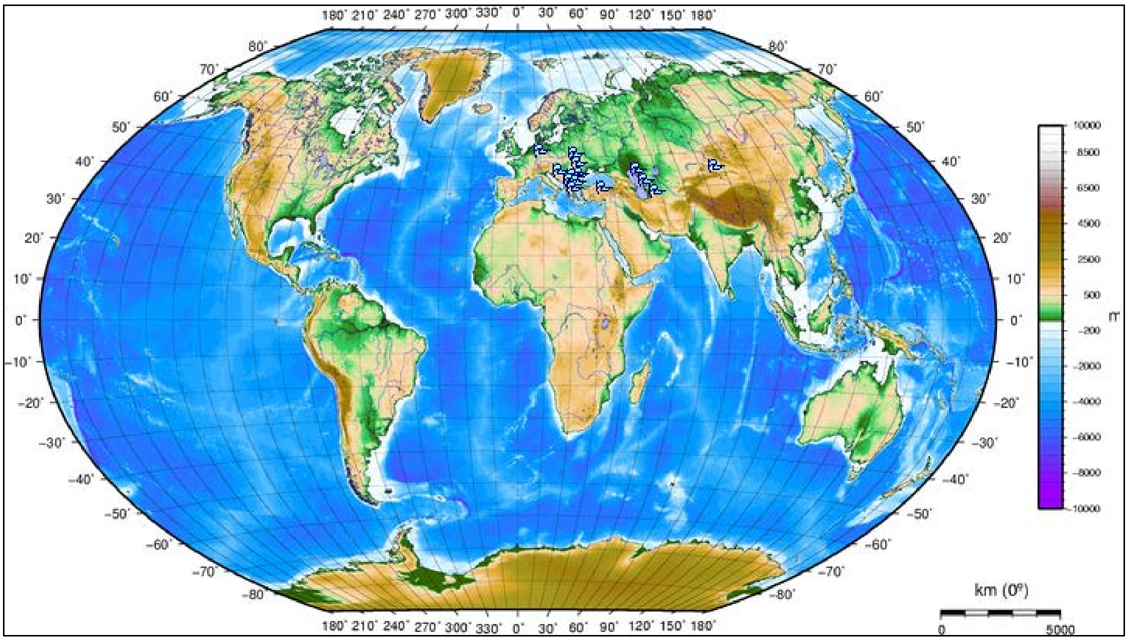
The causes, effects, trends and possibilities of human society to positively intervene to slow down this process or to adapt to it involve a huge variety of aproacess and efforts.

With the fact in mind that these aproaces and efforts shuld be based on genuine scientific understanding, the editors of the *Transylvanian Review of Systematical and Ecological Research* series launch three annual volumes dedicated to the wetlands, volumes resulted mainly as a results of the *Aquatic Biodiversity International Conference*, Sibiu/Romania, 2007-2017.

The therm wetland is used here in the acceptance of the Convention on Wetlands, signed in Ramsar, in 1971, for the conservation and wise use of wetlands and their resources. **Marine/Coastal Wetlands -** Permanent shallow marine waters in most cases less than six metres deep at low tide, includes sea bays and straits; Marine subtidal aquatic beds, includes kelp beds, sea-grass beds, tropical marine meadows; Coral reefs; Rocky marine shores, includes rocky offshore islands, sea cliffs; Sand, shingle or pebble shores, includes sand bars, spits and sandy islets, includes dune systems and humid dune slacks; Estuarine waters, permanent water of estuaries and estuarine systems of deltas; Intertidal mud, sand or salt flats; Intertidal marshes, includes salt marshes, salt meadows, saltings, raised salt marshes, includes tidal brackish and freshwater marshes; Intertidal forested wetlands, includes mangrove swamps, nipah swamps and tidal freshwater swamp forests; Coastal brackish/saline lagoons, brackish to saline lagoons with at least one relatively narrow connection to the sea; Coastal freshwater lagoons, includes freshwater delta lagoons; Karst and other subterranean hydrological systems, marine/coastal. **Inland Wetlands -** Permanent inland deltas; Permanent rivers/streams/creeks, includes waterfalls; Seasonal/intermittent/irregular rivers/streams/creeks; Permanent freshwater lakes (over eight ha), includes large oxbow lakes; Seasonal/intermittent freshwater lakes (over eight ha), includes floodplain lakes; Permanent saline/brackish/alkaline lakes; Seasonal/intermittent saline/brackish/alkaline lakes and flats; Permanent saline/brackish/alkaline marshes/pools; Seasonal/intermittent saline/brackish/alkaline marshes/pools; Permanent freshwater marshes/pools, ponds (below eight ha), marshes and swamps on inorganic soils, with emergent vegetation water-logged for at least most of the growing season; Seasonal/intermittent freshwater marshes/pools on inorganic soils, includes sloughs, potholes, seasonally flooded meadows, sedge marshes; Non-forested peatlands, includes shrub or open bogs, swamps, fens; Alpine wetlands, includes alpine meadows, temporary waters from snowmelt; Tundra wetlands, includes tundra pools, temporary waters from snowmelt; Shrub-dominated wetlands, shrub swamps, shrub-dominated freshwater marshes, shrub carr, alder thicket on inorganic soils; Freshwater, tree-dominated wetlands; includes freshwater swamp forests, seasonally flooded forests, wooded swamps on inorganic soils; Forested peatlands; peatswamp forests; Freshwater springs, oases; Geothermal wetlands; Karst and other subterranean hydrological systems, inland. **Human-made wetlands -** Aquaculture (e. g., fish/shrimp) ponds; Ponds; includes farm ponds, stock ponds, small tanks; (generally below eight ha); Irrigated land, includes irrigation channels and rice fields; Seasonally flooded agricultural land (including intensively managed or grazed wet meadow or pasture); Salt exploitation sites, salt pans, salines, etc.; Water storage areas, reservoirs/barrages/dams/impoundments (generally over eight ha); Excavations; gravel/brick/clay pits; borrow pits, mining pools; Wastewater treatment areas, sewage farms, settling ponds, oxidation basins, etc.; Canals and drainage channels, ditches; Karst and other subterranean hydrological systems, human-made.

The editors of the *Transylvanian Review of Systematical and Ecological Research* started and continue the annual sub-series (*Wetlands Diversity*) as an international scientific debate platform for the wetlands conservation, and not to take in the last moment, some last heavenly “images” of a perishing world …

This 15th volume included variated researches from diverse wetlands around the world.



The subject areas (⚑) for the published studies in this volume.

No doubt that this new data will develop knowledge and understanding of the ecological status of the wetlands and will continue to evolve.

Acknowledgements

The editors would like to express their sincere gratitude to the authors and the scientific reviewers whose work made the appearance of this volume possible.

The Editors

Editorial Office:

“*Lucian Blaga” University of Sibiu, Faculty of Sciences, Department of Ecology and Environment Protection, Dr. Ion Raţiu Street 5-7, Sibiu, Sibiu County, Romania, RO-550012, Angela Curtean-Bănăduc (ad.banaduc@yahoo.com, angela.banaduc@ulbsibiu.ro)*

(ISSN-L 1841 – 7051; online ISSN 2344 – 3219)

SPATIAL DYNAMICS OF SPECIES RICHNESS OF PHYTOPLANKTON OF LAKE BALKHASH IN THE GRADIENT OF ABIOTIC FACTORS

Sophia BARINOVA *, *Elena KRUPA* ** and *Ulrika KADYROVA* ***

* Institute of Evolution, University of Haifa, Mount Carmel, 199 Abba Khoushi Ave., Haifa 3498838, Israel, sophia@evo.haifa.ac.il

** Republican State Enterprise on the Right of Economic Use "Institute of Zoology", Ministry of Education and Science, Science Committee, Almaty, Kazakhstan 050060, elena_krupa@mail.ru

*** "Kazakh Research Institute of Fishery" LLP, 20 Zheltoksan street, Balkhash town, 100300, Republic of Kazakhstan, ylirika@inbox.ru

DOI: 10.1515/trser-2017-0009

KEYWORDS: biodiversity, macrophytes, TDS, biogenes, heavy metals, Kazakhstan.

ABSTRACT

The spatial distribution of species richness of phytoplankton was studied along the Balkhash lake water area during the summer of 2004. With statistically weak connections of phytoplankton with environmental parameters, three-dimensional graphs revealed a complex character of its spatial variability in the gradient of environmental factors. Macrophytes had a stimulating effect on the species richness of planktonic algae. We found a correlation between species richness of Cyanobacteria, Chrysophyta, Euglenophyta and Dinophyta and the ionic composition of water. The important role of temperature was traced. Species richness of algal communities increased with increasing temperature in the gradient of nutrients and correlated with the abundance of macrophytes.

ZUSAMMENFASSUNG: Die räumliche Dynamik der Artenvielfalt des Phytoplanktons im Balkhasch-See/ Kasachstan entlang der Gradienten abiotischer Faktoren.

Die räumliche Verteilung der Artenvielfalt des Phytoplanktons wurde im Sommer 2004 entlang des Balkhash-See-Gebiets untersucht. Bei statistisch schwachen Verbindungen von Phytoplankton mit Umweltparametern zeigten dreidimensionale Graphiken den komplexen Charakter ihrer räumlichen Variabilität im Gradienten der Umweltfaktoren. Makrophyten hatten eine stimulierende Wirkung auf den Artenreichtum der Planktonalgen. Gezeigt wird in vorliegender Arbeit die Korrelation zwischen Artenreichtum von Cyanobakterien, Chrysophyta, Euglenophyta und Dinophyta und der ionischen Zusammensetzung des Wassers. Verfolgt wurde auch die wichtige Rolle des Temperaturfaktors. Der Artenreichtum der Algengemeinschaften nahm mit zunehmender Temperatur entlang des Nährstoffgradienten und in Korrelation mit der Makrophyten-Abundanz zu.

REZUMAT: Dinamica spațială a bogăției speciilor de fitoplancton din lacul Balkhash (Kazahstan) în corelaţie cu gradientul factorilor abiotici.

Distribuția spațială a bogăției speciilor de fitoplancton a fost studiată de-a lungul lacului Balkhash în vara anului 2004. Cu conexiuni slabe statistic ale fitoplanctonului cu parametrii de mediu, graficele tridimensionale au relevat un caracter complex al variabilității spațiale în gradientul factorilor de mediu. Macrofitele au un efect stimulator asupra bogăția speciilor de alge planctonice. Exită o corelație între bogăția de specii de cianobacterii, Chrysophyta, Euglenophyta și Dinophyta și concentraţia ionilor în apă. A fost evidenţiat şi rolul important al temperaturii în structurarea comunităţilor de alge planctonice. Bogăția în specii a comunităţilor de alge creşte odată cu creşterea temperaturii apei, cu cantitatea de nutrienţi din apă şi se corelează cu abundenţa macrofitelor.

INTRODUCTION

Lake Balkhash (Figs. 1-3) (south-east of Kazakhstan) runs from the south-west to the northeast. At surface level, it is 342 m is 614 km long and covers an area of approximately 16.4 thousand km^2 (Kudekov, 2002). The Uzun-Aral Strait divides the lake into two parts – the Western and Eastern Balkhash. The food is provided by the rivers Ily, Karatal, Aksu, Lepsy, originating in the mountains of the Tien Shan in the zone of glaciers and flows into the lake from the south. The peculiarity of Balkhash is the growth of the total content of dissolved salts along the longitudinal axis, in connection with which its water area is divided into 8 hydrochemical regions (Fig. 1). (Tarasov, 1961)

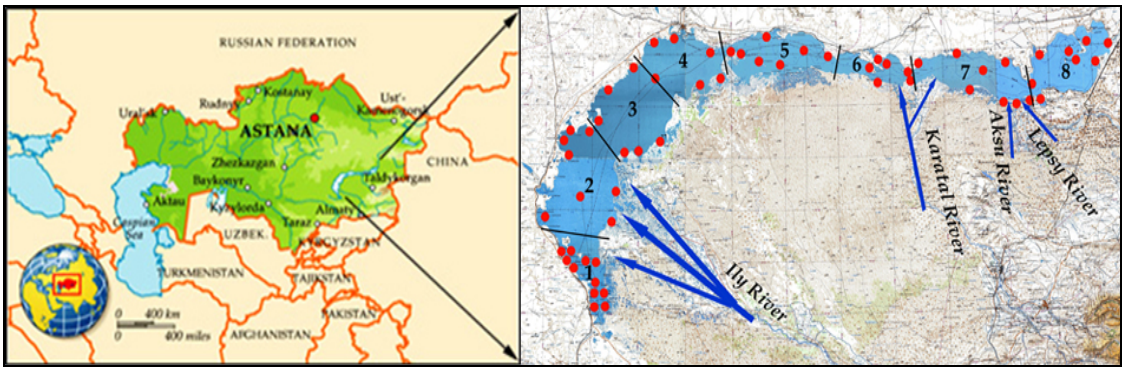


Figure 1: Map sampling stations at Lake Balkhash, 2004;
blue arrow are the river water input,
1-8 are hydrochemical regions.

The average value of TDS in different years varies from 0.60-1.20 g/dm^3 in the west to 4.04-5.60 g/dm^3 in the east (Kudekov, 2002). The lake is located in an area with large deposits of polymetallic ores (Mazurov, 2005), which are responsible for the increased background levels of heavy metals in the water and soil. Technogenic pollution of Balkhash and adjacent territories is due to industrial extraction and processing of minerals (Samakova, 2002). The significant heterogeneity of abiotic factors affects the spatial distribution of phytoplankton as the primary trophic level of any aquatic ecosystem.

Systematic studies of phytoplankton in Lake Balkhash have been carried out since 1971. The focus has been the study of the species composition of planktonic and benthic algae (Akhmetova, 1986), the identification of dominant species, and the assessment of the level of feeding in the hydrochemical regions of the lake (Abrosov, 1973; Vorobyeva et al., 1982; Fokina, 1982; Ponomareva et al., 2005) in relation to climatic factors (Krupa et al., 2014).

The aim of this work is to study the spatial distribution of species richness of phytoplankton in the water area of Lake Balkhash in the gradient of hydrophysical, hydrochemical and toxicological factors.

Figure 2: Lake Balkhash.

Figure 3: Lake Balkhash.

MATERIAL AND METHODS

The research of phytoplankton and total dissolved solids (TDS) content was carried out by means of a grid of 58 stations (Fig. 1) in June and July of 2004. The measures of the temperature and pH values of the surface water layers were taken in the field environment. Water transparency was measured with a Secchi disk. Coordinate referencing of the stations was done by Garmin eTrex GPS-navigator. The samples for heavy metals were fixed in the site by adding nitric acid; samples for biogenes were fixed with chloroform. All collected samples were transported to the lab in an icebox.

Conventional methods of chemical analysis of water were used (Semenova, 1977; Fomin, 1995). Water samples were analyzed in three – four replications. The error of estimate for major ions in the water was 0.5-5.0%, depending on the analyte. Heavy metal measuring was performed by mass spectrometry with inductively coupled plasma by using Agilent 7500 A manufactured by Agilent Technologies, USA (National Standard RK ISO 17294-2-2006). The device allows for the detection of the various chemical elements in complex matrices, including those in the sea and grey water and in the biological objects in micro-trace quantities. Abundance Sensitivity of Agilent 7500 A: Low Mass < 5×10^{-7}, High Mass < 1×10^{-7}.

For the processing of phytoplankton samples, the settling method was used (Kiselev, 1956). Species identification of planktonic algae was performed by using determinants for relevant divisions (Zabelina et al., 1951; Gollerbach et al., 1953; Popova, 1955; Palamar-Mordvintseva, 1982; Moshkova and Gollerbach, 1986). The statistical methods were implemented in the GRAPHS program (Novakovsky, 2004) used for the comparative floristics and Statistica 12.0 for nonparametric correlation analysis, spatial mapping innovative approach and surface plots' construction in analysis of biological and environmental variables' relationship.

The integral index of aquatic ecosystem sustainability (WESI) was constructed on results of our studies (Barinova et al., 2006). It is based on the water quality ranges as determined by saprobity indices S and nitrate (or phosphates) concentrations. The resulting WESI index reflects the self-purification capacities of the aquatic ecosystem. At WESI ≥ 1, the photosynthetic level is positively correlated with the level of nitrate concentration. At WESI < 1, photosynthesis is suppressed (presumably due to a toxic disturbance) if the water body is slightly contaminated by toxicants.

RESULTS

The Western and Eastern Balkhash significantly differ in their characteristics, as can be seen in table 1. The western part of the lake is shallow, with less transparency, lower water pH, and less spread of hard macrophytes, but greater water coverage by soft macrophytes. The deeper and narrower eastern part of the water area is overgrown with hard and soft macrophytes to approximately the same degree. The hard macrophytes are represented by *Phragmites australis* (Cav.) Trin. ex Steud., *Schoenoplectus lacustris* (L.) Palla; the soft macrophytes by *Potamogeton críspus* L., *Stuckenia pectinata* (L.) Böerner, *Nymphaea* sp., *Nuphar* sp., *Myriophyllum spicátum* L., *Ceratophyllum* sp., as well as the charophytic macroalgae *Chara tomentosa* Linnaeus and *Nitellopsis obtusa* (N.A.Desvaux) J.Groves. Macrophytes develop mainly near the southern and southeastern shores of the lake, in shallow-water zones under the influence of river flow (Fig. 2).

Table 1: Hydrophysical and morphometric characteristics of the lake; Balkhash, summer 2004.

Variable	Whole Balkhash	Western Balkhash	Eastern Balkhash
Temperature, °C	24.14±0.14	23.49±0.16	24.77±0.14
Depth, m	5.58±0.49	4.34±0.28	6.86±0.81
Transparency, m	1.13±0.15	0.53±0.02	1.74±0.25
Macrophytes Hard, %	20.05±3.03	18.03±3.00	22.14±5.36
Macrophytes Soft, %	25.18±2.68	28.28±3.29	22.00±4.24
pH	8.63±0.04	8.52±0.02	8.74±0.09

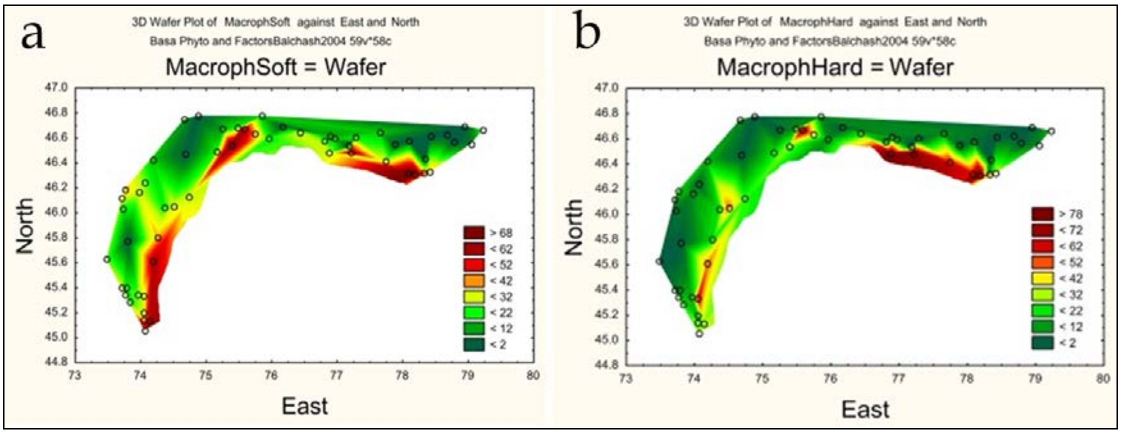


Figure 2: Distribution of soft and hard macrophytes along Lake Balkhash, summer 2004.

The TDS of the East Balkhash water was statistically significantly higher than in the western part during the summer of 2004 (Tab. 2). The ionic composition was dominated by sulphates and sodium. The total content of dissolved solids in the water increased in the direction from west to east, with a simultaneous increase in the concentration of all ions except calcium. Higher concentrations of nitrates, nitrites, total phosphorus, iron, zinc, and copper are found in Western Balkhash water. Phosphates, readily oxidized organic matter, cadmium, lead, nickel and cobalt were present in large quantities in the water of the eastern part of the water area. The content of ammonia and silicon did not differ in the two parts of the lake.

The revealed phytoplankton was represented by 91 species, of which greens 29, diatoms 26, cyanobacteria 21, charophytes 4, euglenophytes 4, dinophytes 3, and chrysophytes 1. No more than 7-15 species were represented in algal communities in most of the water area, as a general rule. In Western Balkhash, where Chlorophyta prevailed (Fig. 3a), the number of species of plankton algae (74) was higher than in Eastern Balkhash (69), where the basis of species richness was formed by Bacillariophyta. In Western Balkhash, the most frequently encountered dinophyte *Peridinium* sp., diatom *Cyclotella meneghiniana* Kützing, and euglenophyte *Trachelomonas* sp. In Eastern Balkhash, in addition to the last two species, diatom *Navicula* sp., green alga *Franceia* sp., and cyanobacteria species *Snowella lacustris* (Chodat) Komárek and Hindák, *Gomphosphaeria aponina* Kützing and *Gloeocapsa* sp. were represented. Species composition of algae underwent changes in the hydrochemical regions of the lake (Fig. 3c), with an increase in the species richness of cyanobacteria and a decrease in the number of species of euglenoic, chrysophytes and dinophytes, as a response to changes in the ionic composition of the lake water.

The hydrochemical regions' communities of the lake were divided into three clusters at the similarity level of the species composition of planktonic algae more than 50% (Fig. 3a). The first cluster united the phytoplankton species of the greater part of Western Balkhash (hydrochemical regions 1, 2, and 4). The second cluster included communities of East Balkhash, except for the 7th region, and algal communities of the 3rd hydrochemical region of the western part. An area with a unique species composition of planktonic algae was the 7 hydrochemical region, which separated into a different cluster.

Table 2: Total Dissolved Solids (TDS), hydrochemical water variables and toxicants of Balkhash Lake, summer 2004; all Variables in mg dm^{-3}, oxidability in mgO_2 dm^{-3}.

*Variable	Whole Balkhash	Western Balkhash	Eastern Balkhash
Ca	40.4±2.5	48.8±4.4	32.4±1.3
Mg	67.4±6.6	35.0±2.2	98.4±8.4
Na+K	454.8±53.8	202.6±22.9	695.6±71.8
HCO_3	415.1±31.6	254.3±11.0	568.6±38.6
SO_4	834.9±93.1	369.1±344	1279.5±116.2
Cl	473.3±58.9	170.5±18.6	762.4±71.3
TDS	2286.0±236.0	1080.3±83.4	3436.8±286.7
Na	448.9±51.1	193.8±2.2	692.4±63.3
K	30.4±4.1	8.8±1.0	51.0±4.9
K/Na	0.063±0.004	0.046±0.0009	0.080±0.005
K/Ca	0.978±0.161	0.196±0.022	1.725±0.215
Mg/Ca	2.021±0.256	0.749±0.039	3.235±0.331
NH_4	0.102±0.017	0.103±0.013	0.102±0.033
NO_3	0.945±0.244	1.379±0.410	0.492±0.226
NO_2	0.042±0.011	0.060±0.013	0.024±0.016
P-PO_4	0.017±0.004	0.011±0.003	0.023±0.007
P_{tot}	0.064±0.001	0.106±0.012	0.021±0.007
Fe	0.032±0.005	0.056±0.006	0.010±0.0006
Si	5.49±0.15	5.20±0.22	5.76±0.20
Oxidability	7.39±0.46	5.07±0.40	9.13±0.45
Zn	0.028±0.009	0.039±0.018	0.017±0.002
Cu	0.018±0.003	0.022±0.005	0.013±0.002
Cd	0.0036±0.0002	0.0028±0.0002	0.0044±0.0002
Pb	0.034±0.003	0.021±0.002	0.047±0.004
Ni	0.039±0.001	0.037±0.002	0.042±0.002
Co	0.013±0.001	0.010±0.0005	0.017±0.001

The dendrite of species composition similarity, constructed by the Ward method, singled out the communities of the 3rd hydrochemical region as a core, which include the largest number of species from other parts of the water area (Fig. 3b). Phytoplankton of the 3rd region, in turn, were close to the algal communities of the 1st region, the latter being the floristic core for the entire lake.

The nonparametric correlation analysis revealed the presence of predominantly weak and moderate in strength relationships between abiotic factors and spatial dynamics of algae species richness (Tab. 3). The total number of species in algal communities increased with macrophytes growing in shallow water areas, with an increase in K/Na values and a decrease in the concentrations of nitrites, copper and total toxic pollution expressed by the WESI index. The dynamics of the species number in the gradient of environmental parameters was generally similar, with some additional features. In addition to the factors listed above, table 3 show that Chrysophyta was affected by a reduction in the species number in response to an increase in cobalt and lead, Bacillariophyta for total phosphorus, and Cyanobacteria for calcium and iron increasing. Euglenophyta had a positive relationship with calcium, but showed a negative dependence on chloride. Cyanobacteria, in contrary, showed a negative relationship with nitrites, nitrates, total phosphorus, and calcium and a positive relationship with all other ions and total dissolved solids.The tendency with statistically weak connections is an increase in the total number of species of algae of macrophytes in shallow heated waters.

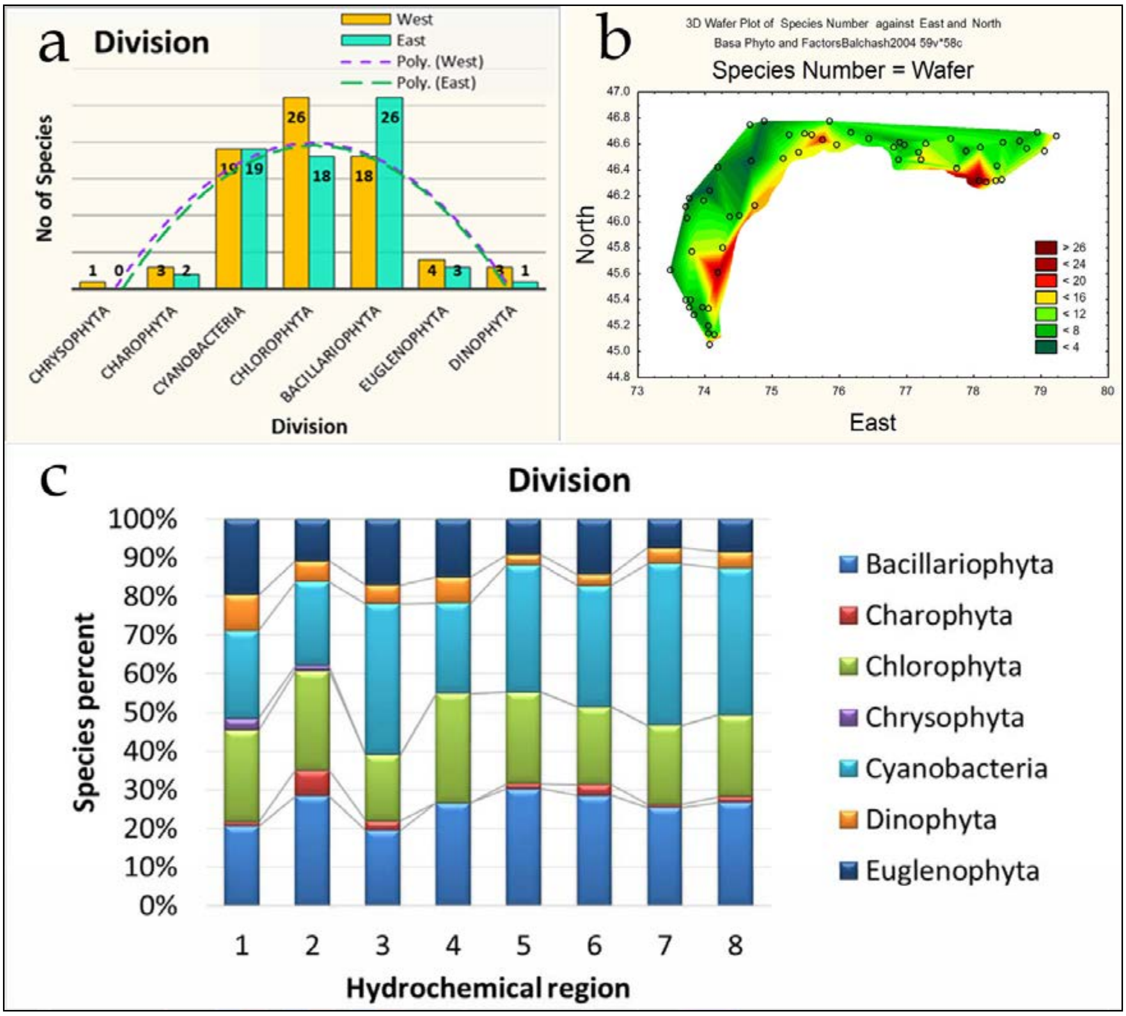


Figure 3: Distribution of phytoplankton species richness taxonomic Division in the west and east part (a), water surface (b), hydrochemical regions (c) of Lake Balkhash, summer 2004.

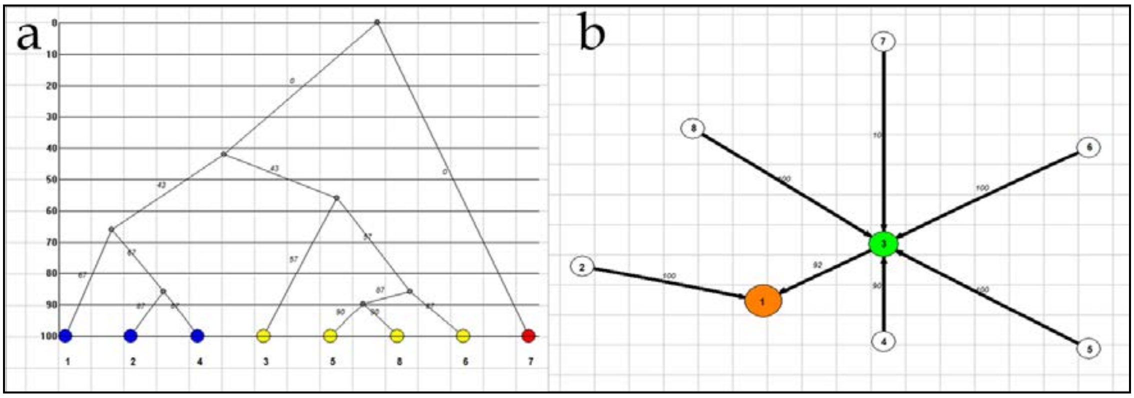


Figure 4: Dendrogram and dendrite similarities in species composition of phytoplankton in the hydrochemical regions of Lake Balkhash, summer 2004.

The maps showed that local outbreaks of the species richness of phytoplankton (Fig. 3b) were confined to the southeastern and southern shores of the lake, ie, to the areas of the aquatic area that overgrew macrophytes (Fig. 2). The tendency to increase the species richness of algal communities in connection with macrophytes was most clearly seen on 3d Surface Plots. With an almost linear increase in the number of algal species in the gradient of temperature and overgrowing by soft macrophytes (Fig. 5e), the connection with hard macrophytes was nonlinear (Fig. 5f). The tendency to increase the number of species of plankton algae with decreasing depth and rising water temperature (Fig. 5b) indirectly reflected the connection with macrophytes, confined to shallow, well-warmed areas of the lake. The leading role of the temperature factor in the formation of the species richness of algal communities was also traced in combination with the pH value and water transparency (Figs. 5c, d).

The relationship between the species richness of phytoplankton and TDS and the chemical composition of water was complex (Fig. 6), with general trends in the variability of the number of species depending on specified environmental parameters. Species richness of algae almost linearly increased in the gradient of calcium concentrations and temperature (Fig. 6a). The algal communities were represented by the maximum number of species at high temperatures and low concentrations of potassium (Fig. 6b), sodium (Fig. 6c), sulphates (Fig. 6f), chlorides (Fig. 6g), TDS (Fig. 6h) and the entire range of magnesium concentrations (Fig. 6d). The variability of the number of species under low-temperature conditions was non-linear in the gradient of potassium, sodium, and TDS, with a second, less pronounced peak in the gradient of carbonates and chlorides.

The algal communities of the lake showed a relative linear increase in the species number at maximum temperatures in the gradient of iron, ammonium, and nitrates (Figs. 7a, b, d). The relationship of the communities species richness with nitrites (Fig. 7c), the content of the easily oxidizable organic matter (Fig. 7e) and, partialy, with the total phosphorus (Fig. 7g) was nonlinear, and with phosphate was absent (Fig. 7f). The maximum of species richness is in the heated areas of the water surface, but with a low content of silicon in water (Fig. 7h).

The 3d Surface Plots revealed a rather complex character of the variability of the total number of algal species in the gradient of heavy metal concentrations (Fig. 8). The greatest phytoplankton species richness was observed in the warmest areas of the water surface where there were high concentrations of cobalt, copper, and nickel (Figs. 8b,c,d) and in the entire range of lead concentration (Fig. 8e). The effect of cadmium (Fig. 8a) and zinc (Fig. 8f) on the number of planktonic species of algae was manifested only locally.

Table 3: The coefficients of Spearman Rank Order Correlations between the abiotic characteristics and the phytoplankton in Balkhash Lake, summer 2004, $p < 0.05$.

Paired variables	Spearman Rank Order Correlations	Paired variables	Spearman Rank Order Correlations	Paired variables	Spearman Rank Order Correlations
Bacillariophyta – Depth	-0.331	Cyanobacteria – HCO_3	0.501	Dinophyta – Na	-0.353
Bacillariophyta – K/Na	0.314	Cyanobacteria – K	0.472	Dinophyta – SO_4	-0.315
Bacillariophyta – MacrophHard	0.338	Cyanobacteria – K/Ca	0.496	Dinophyta – TDS	-0.342
Bacillariophyta – MacrophSoft	0.356	Cyanobacteria – K/Na	0.513	Euglenophyta – Ca	0.375
Bacillariophyta – NO_2	-0.397	Cyanobacteria – Mg	0.453	Euglenophyta – Cl	-0.308
Bacillariophyta – Ptot	-0.319	Cyanobacteria – Na	0.458	Euglenophyta – Depth	-0.420
Bacillariophyta – Temperature	0.311	Cyanobacteria – Na+K	0.511	Euglenophyta – MacrophSoft	0.337
Charophyta – WESI P_PO_4	0.309	Cyanobacteria – SO_4	0.413	Euglenophyta – Na	-0.303
Charophyta – MacrophSoft	0.327	Cyanobacteria – TDS	0.469	Euglenophyta – Na+K	-0.377
Chlorophyta – MacrophHard	0.337	Cyanobacteria – NO_2	-0.535	Euglenophyta – NH_4	0.424
Chlorophyta – WESI P_PO_4	0.600	Cyanobacteria – NO_3	-0.349	Species Number – Depth	-0.452
Chrysophyta – Cl	-0.348	Cyanobacteria – Ptot	-0.467	Species Number – MacrophHard	0.489
Chrysophyta – Co	-0.409	Cyanobacteria – Temperature	0.465	Species Number – MacrophSoft	0.420
Chrysophyta – Fe	0.390	Cyanobacteria – Transparency	0.368	Species Number – Temperature	0.349
Chrysophyta – Pb	-0.431	Cyanobacteria – WESI NO_3	0.508	Species Number – NO_2	-0.407
Cyanobacteria – Ca	-0.465	Cyanobacteria – WESI NO_3	0.508	Species Number – Ptot	-0.346
Cyanobacteria – Cd	0.498	Dinophyta – Cl	-0.366	Species Number – K/Na	0.466
Cyanobacteria – Cl	0.481	Dinophyta – HCO_3	-0.356	Species Number – Cu	-0.405
Cyanobacteria – Cu	-0.552	Dinophyta – K	-0.363	Species Number – WE SI P_PO_4	0.453
Cyanobacteria – Fe	-0.591	Dinophyta – K/Ca	-0.323	Species Number – WESI NO_3	0.424

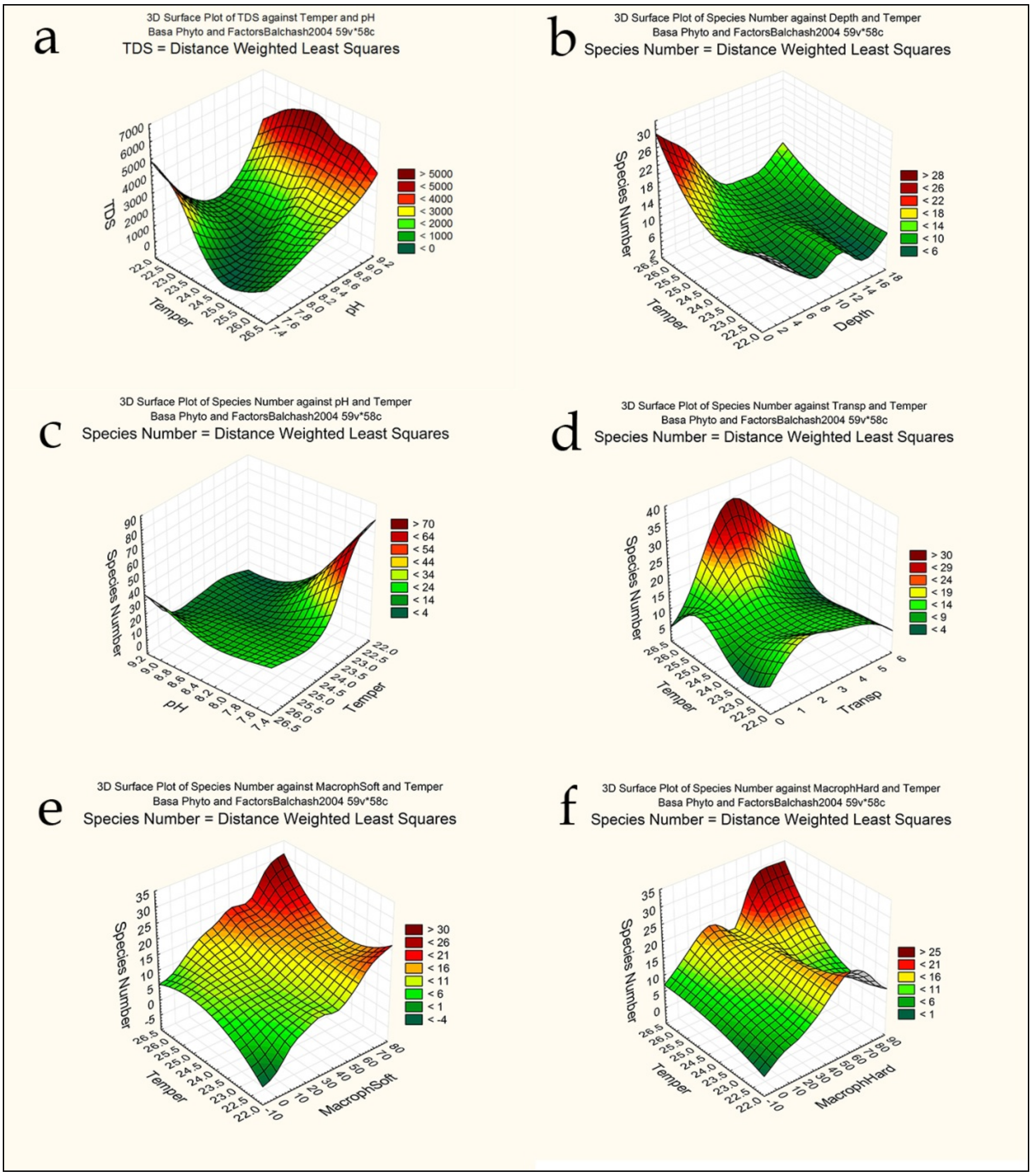


Figure 5: Distribution of species richness of phytoplankton depending on the hydrophysical factors of Lake Balkhash, summer 2004.

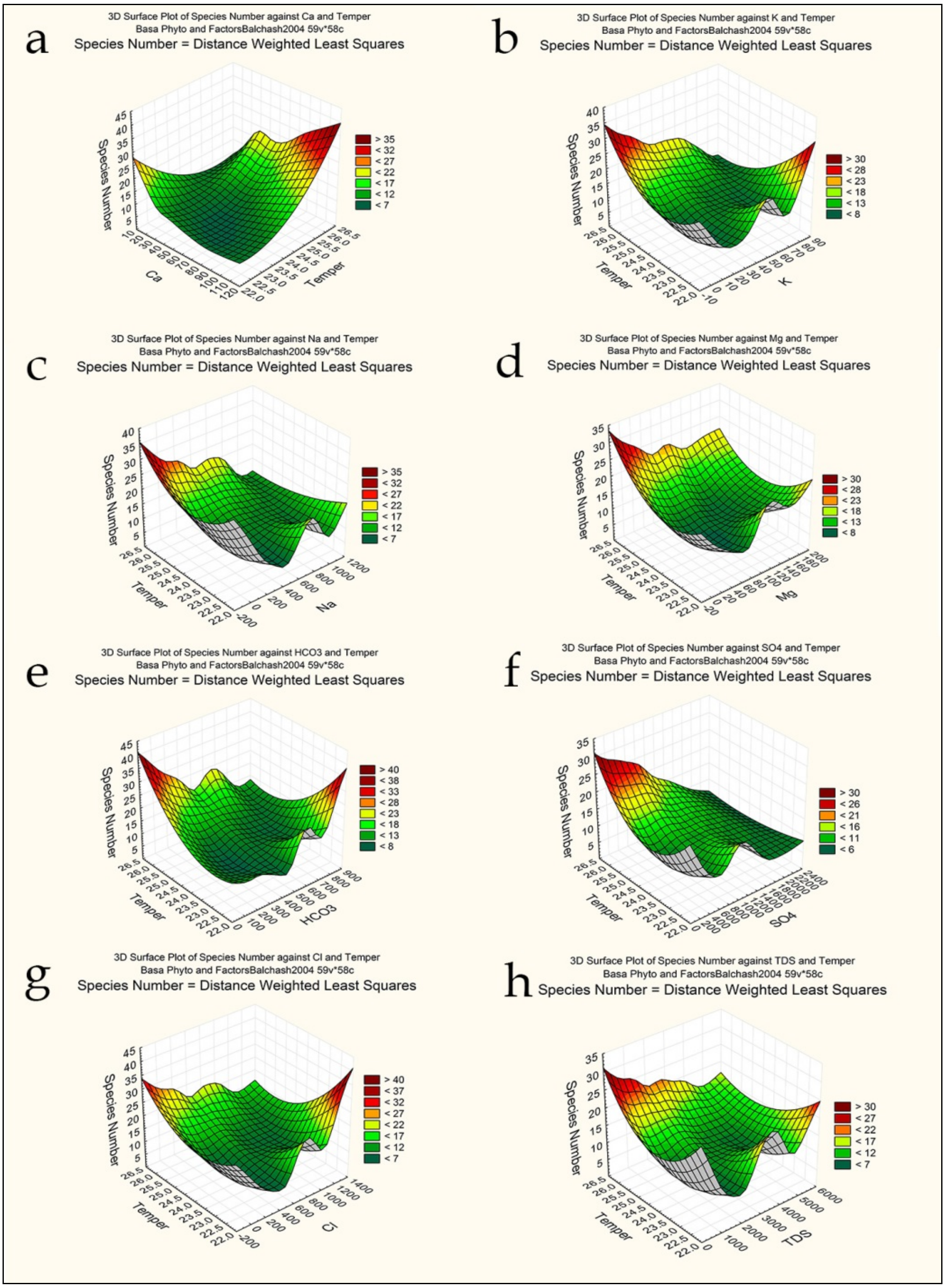


Figure 6: Spatial distribution of species richness of phytoplankton depending on the ion composition and total dissolved solids (TDS) in the water of Lake Balkhash, summer 2004.

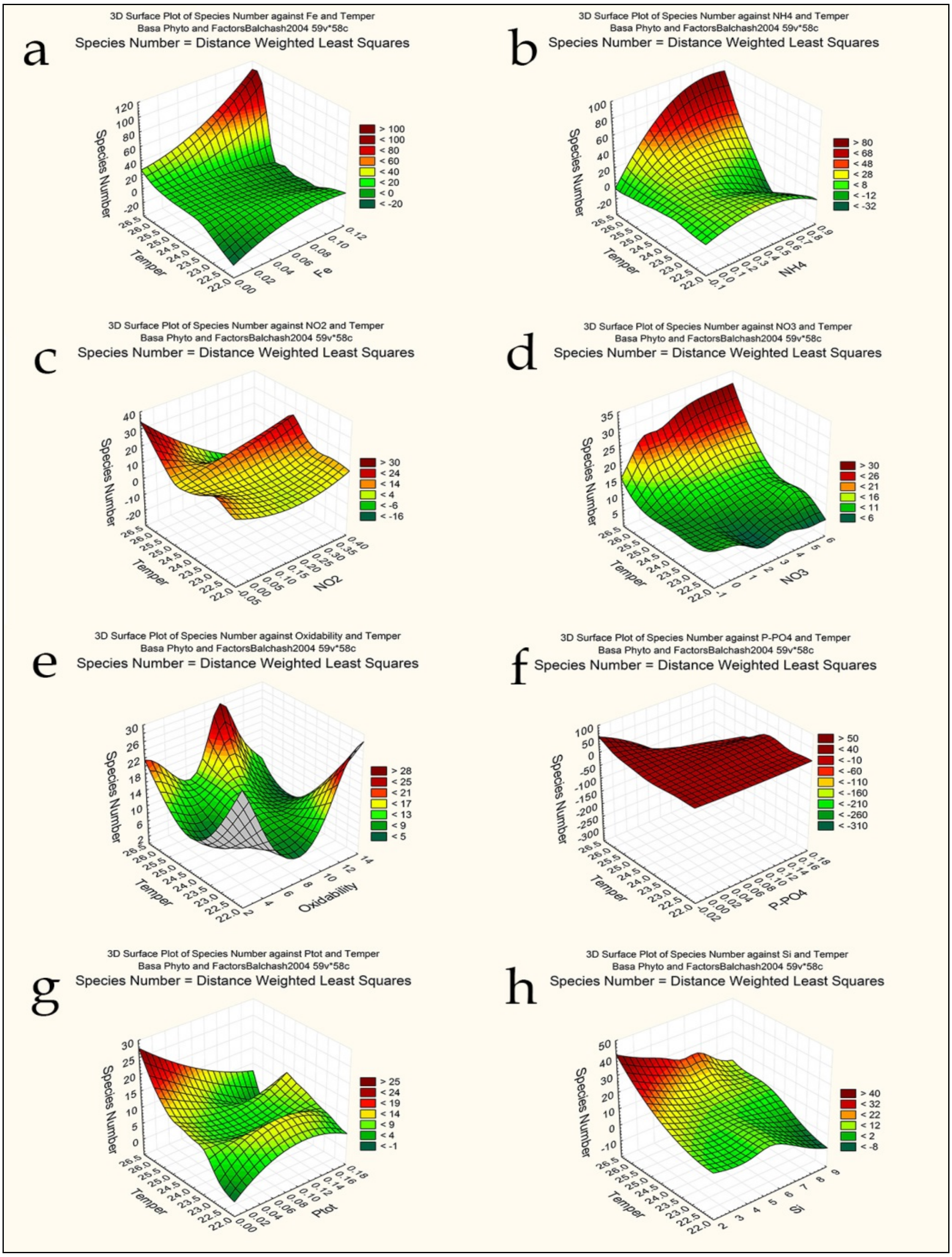


Figure 7: Spatial distribution of species richness of phytoplankton depending on the content of biogenic elements and easily oxidized organic matter in the water of Lake Balkhash, summer 2004.

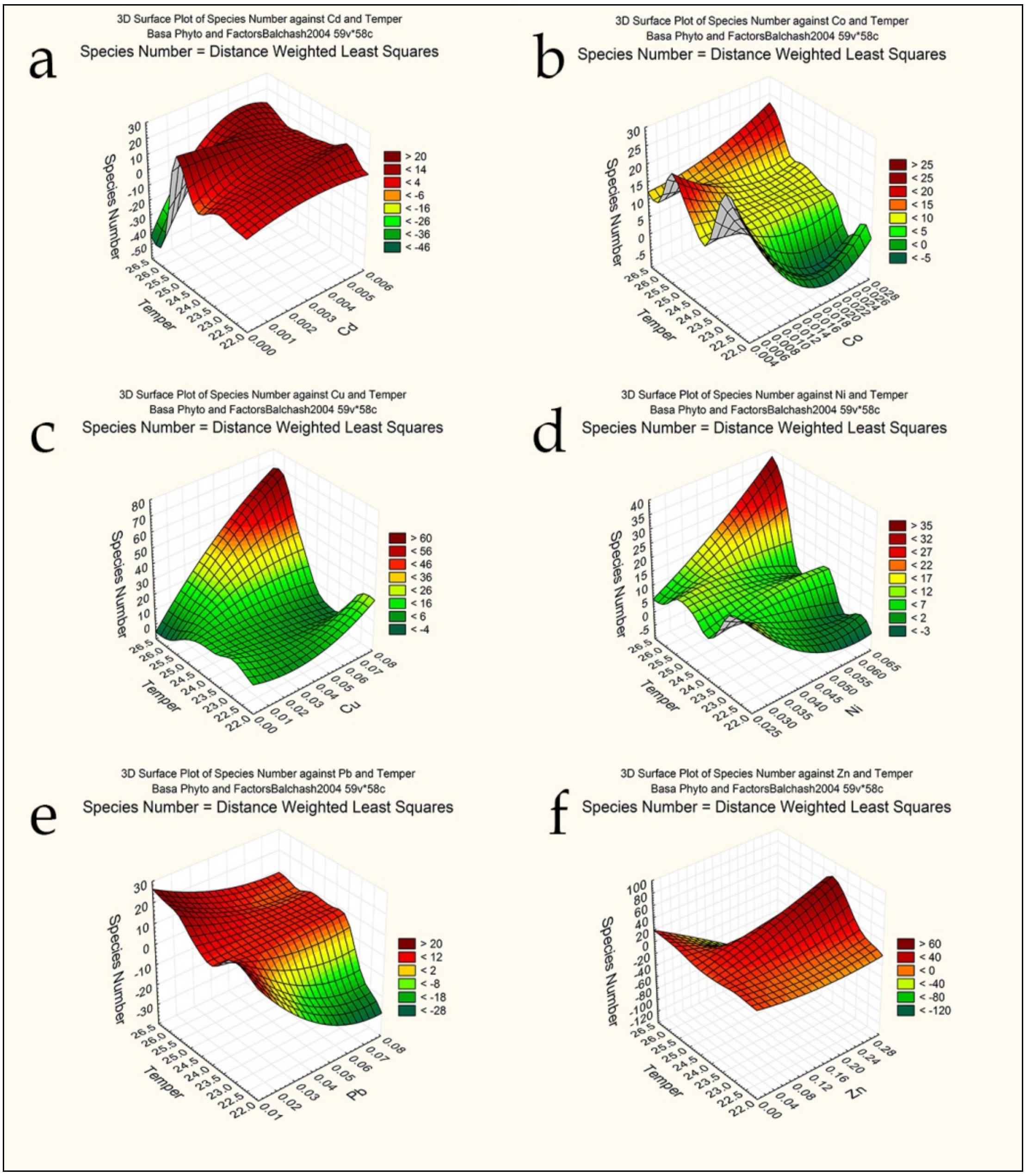
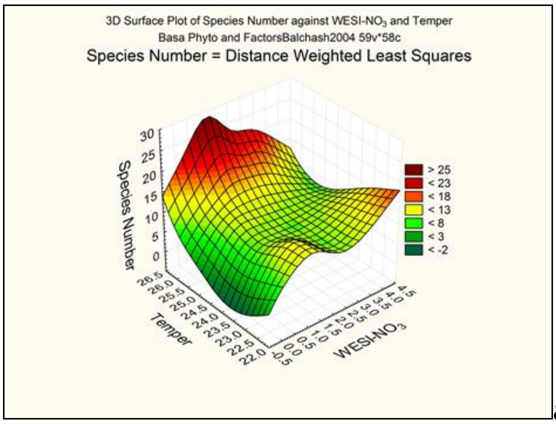
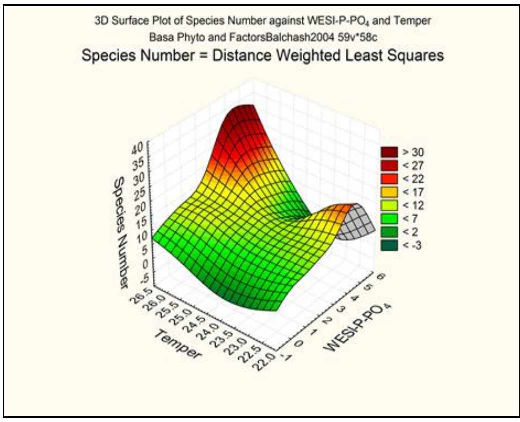


Figure 8: Spatial distribution of species richness of phytoplankton depending on the content of biogenic elements and easily oxidized organic matter in the water of Lake Balkhash, summer 2004.



a.



b.

Figure 9a, b: Spatial distribution of species richness of phytoplankton in the gradient of total toxic pollution of Lake Balkhash in summer 2004.

Reduction of the total toxic pollution, expressed by the WESI-NO_3 index, stimulated the phytoplankton growth in species richness in the high temperature zone and, to a lesser extent, in the less heated areas of the lake (Fig. 9a). The most richest algal communities were represented at the maximum values of WESI-P-PO_4 in the high-temperature zone also (Fig. 9b).

DISCUSSION

The phytoplankton of Lake Balkhash was comprised of 91 species, with a variety of green, diatoms and cyanobacteria, in the summer of 2004. The species composition of the phytoplankton underwent changes in the hydrochemical regions of the lake, with an increase in the cyanobacteria species number and a decrease in the number of species of euglenoids, chrysophytes and dinophytes algae, as a response to a change in the ionic composition of water in the direction from west to east.

The species richness of phytoplankton communities was clearly traced in the salt gradient for water bodies in some regions (Barinova et al., 2009, 2011, 2015; Klymiuk et al., 2014; Bilous et al., 2016). The non-parametric correlation analysis results and 3d Surface Plots revealed a more complex character of the relationship of species richness of algae with hydrochemical conditions in Lake Balkhash. The lack of a clear connection between TDS and the total species richness of phytoplankton is associated with a different reaction of algae to the salt composition of the water. The number of species of Cyanobacteria and Chrysophyta increased in the gradient of salinity or concentrations of individual ions positively associated with TDS, whereas the species richness of Euglenophyta and Dinophyta was decreased. The chemical composition of the water did not exert any significant influence on Charophyta and Chlorophyta species. Other environmental parameters also contributed to the variability of the phytoplankton species richness, the most important of which were the temperature and overgrowing of the water area by macrophytes. Spatial distribution maps and 3d Surface Plots revealed an increase in the phytoplankton species richness with statistically weak connections in warmed shallow zones and macrophyte overgrowth. As shown by the analysis of reference data, a lower diversity of phytoplankton (Muylaert et al., 2010) is observed in the thickets of macrophytes in the humid zone reservoirs, as compared to the open areas, due to the high abundance of large plankton invertebrates eating algae (Pieczy'nska et al., 1999). The previously noted positive relationship between macrophytes and the number of phytoplankton species (Barinova and Krupa, 2017) is due to the fact that on the overgrowing areas of the arid zone water bodies, the role of small species in zooplankton, mainly rotifers, (Krupa, 2012) are increased, which does not have a controlling effect on algae. Along with the temperature, phosphorus and nitrogen are the most important elements controlling the development of planktonic algae (Goncharov, 2007; Tolotti et al., 2012; Mingli et al., 2014). As the temperature and loading of nutrients rise, a gradual loss of the biological diversity of the aquatic ecosystem takes place (Elliott et al., 2006).

The important role of temperature, despite the relatively small amplitude of its oscillations, was traced when assessing the influence of all environmental parameters on the planktonic algae of Balkhash. The same environmental indicator could have a different effect on the species richness of algae in the zone of relatively low (22.5-23.0°C) and high temperatures (25.0-26.0°C). Algal communities showed an almost linear increase in the number of species in the gradient of iron, ammonium, nitrates, and calcium at the maximum temperature. Unfavourable conditions for algae were formed with an increase in TDS, high concentrations of potassium, sodium, sulphates, and chlorides in warmed areas of the lake, while magnesium did not affect the species richness of phytoplankton. The variability of the number of species under relatively low temperatures was nonlinear in the gradient of potassium, sodium, and TDS, and had a second, less pronounced peak in the gradient of carbonates and chlorides. We did not find a direct correlation of the species richness in communities with phosphates, but the positive values of the WESI-P-PO_4 index indicated a sufficient amount of phosphate in the lake. Thus, the dual nature of the relationship between species richness in algal communities and environmental parameters was traced, and it increased with increasing temperature in the nutrient gradient, but under conditions of minimum values of TDS and concentrations of the main ions. The positive effect of temperature on the number of species of planktonic algae is revealed with even a small gradient of this factor in the conditions of Balkhash. Taking into account the literature data (Elliott et al., 2006), with a further increase in water temperature, one can expect a decrease in the species richness of phytoplankton that is an important prognostic conclusion in the future as it relates to global warming.

While maintaining the leading role of the temperature factor, 3d Surface Plots have revealed a complex, but weakly expressed, character of the dependence of the species richness of algae with heavy metals. This may be due to the increased content of dissolved solids in Balkhash water, which, along with the alkaline reaction of water, contribute to reducing the toxic properties of heavy metals by finding the latter in an insoluble form (Nikanorov and Zhulidov, 1991). The obtained results with the new methods used showed that statistically significant, but weak, relationships can indicate a complex character of the spatial variability of biological variables in the gradient of environment factors. One of the methods for detecting such nonlinear dependences is 3d Surface Plots construction, the use of which has made it possible to describe the variability of the phytoplankton species richness quite realistically with the hydrophysical, hydrochemical and toxicological variables of Lake Balkhash.

CONCLUSIONS

Nonparametric correlation analysis revealed a weakly, but statistically significant, relationship between the phytoplankton species richness and the environmental variables of Lake Balkhash. This is due to the complex nature of the spatial variability of phytoplankton in the gradient of environmental factors, which is clearly demonstrated by three-dimensional graphs. The stimulating effect of macrophytes on the species richness of plankton algae is shown. The absence of a connection between the total species richness of phytoplankton and TDS is associated with a different reaction of algae to the salt composition of the water. The number of species of Cyanobacteria and Chrysophyta was increased, whereas Euglenophyta and Dinophyta were decreased in the gradient of salinity and concentrations of individual ions. The chemical composition of the water had no significant effect on Charophyta and Chlorophyta species. The important role of the temperature factor was traced when we assessed the influence of all environmental variables on the planktonic algae of Balkhash. The same environmental variable caused different variability in the species richness of algae in the low (22.5-23.0°C) and high temperature (25.0-26.0°C) ranges. At maximum temperatures, the number of species of algae increased linearly in the gradient of iron, ammonium, nitrates, calcium, and decreased with TDS, high concentrations of potassium, sodium, sulphates, and chlorides, and throughout the magnesium content range. The variability of the number of species was non-linear in the gradient of potassium, sodium, and TDS, and had a second, less pronounced peak in the gradient of carbonates and chlorides under low-temperature conditions. Thus, the dual nature of the relationship between the species richness of algal communities and environmental variables was traced in its gradient in environment as species richness increased with increasing temperature in the nutrient gradient, but with a decrease in TDS and the main ions in water. With an increased content of heavy metals in Lake Balkhash, they did not have a significant effect on the species richness of algae. This is due to the increased content of dissolved solids in Balkhash and the alkaline reaction of water.

ACKNOWLEDGEMENTS

The work was carried out partly under the project № 1846/ГФ4 Г.2015-Г2016 for Committee of Science, Ministry of Education and Science, Republic of Kazakhstan "Development of the methods for controlling the ecological state of water bodies in Kazakhstan", "Study of the modern hydroecological state of fishery water reservoirs and development of biological justifications for the purposefulness and priority of fishery reclamation for conservation and enhancement of the fishery potential of the reservoir" as well as partly supported by the Israeli Ministry of Absorption.

REFERENCES

1. Abrosov V. N., 1973 – Lake Balkhash, Leningrad, Russia, *Science*, 181. (in Russian)
2. Akhmetova N. I., 1986 – The abstract of the dissertation of the candidate of biological sciences, Leningrad, Russia: Botanical Institute named after N. L. Komarova, 25. (in Russian)
3. Barinova S. and Krupa E., 2017 – Bioindication of Ecological State and Water Quality by Phytoplankton in the Shardara Reservoir, Kazakhstan, *Environment and Ecology Research,* 5, 73 - 92.
4. Barinova S. S., Medvedeva L. A. and Anissimova O. V., 2006 – Diversity of algal indicators in environmental assessment, Tel Aviv: Pilies Studio, 498. (in Russian)
5. Barinova S. S., Klochenko P. D. and Belous Y. P., 2015 – Algae as Indicators of the Ecological State of Water Bodies: Methods and Prospects, *Hydrobiological Journal,* 51, 6, 3-21.
6. Barinova S. S., Bragina T. M. and Nevo E., 2009 – Algal species diversity of arid region lakes in Kazakhstan and Israel, *Community Ecology,* 10, 1, 7-16.
7. Barinova S. S., Nevo E. and Bragina T. M., 2011 – Ecological assessment of wetland ecosystems of northern Kazakhstan on the basis of hydrochemistry and algal biodiversity, *Acta Botanica Croatica,* 70, 2, 215-244.
8. Bilous O. P., Barinova S. S., Ivanova N. O. and Huliaieva O. A., 2016 – The use of phytoplankton as an indicator of internal hydrodynamics of a large seaside reservoir-case of the Sasyk Reservoir, Ukraine, *Ecohydrology and Hydrobiology,* 16, 160-174.
9. Elliott J. A., Jones I. D. and Thackeray S. J., 2006 – Testing the sensitivity of phytoplankton communities to changes in water temperature and nutrient load, in a temperate lake, *Hydrobiologia*, 559, 401-411.
10. Fokina A. S., 1982 – The present state of phytoplankton in the Balkhash Lake. In: Forecast of the integrated and rational use of natural resources, their protection and prospects for the development of the productive forces of the Lake Basin, Balkhash in the period until 1990-2000, Part 2, Alma-Ata, Kazakhstan: Science of KazSSR, 137-140. (in Russian)
11. Fomin G. S., 1995 – Water. Control of chemical, bacterial and radiation safety according to international standards, Moscow, Russia: NGO “Alternative”, 618. (in Russian)
12. Gollerbach M. M., Kossinskaya E. K. and Polyansky V. I., 1953 – Key to freshwater algae of USSR, Vol. 2, Blue-green algae, Moscow, Russia: Soviet Science, 654. (in Russian)
13. Goncharov A. V., 2007 – Comparison of Reservoirs in the Moskva–Vazuza Water System in Terms of Phytoplankton Abundance and Eutrophication Degree, *Water Resources,* 34, 1 70-74.
14. Kiselev I. A., 1956 – Methods of study of plankton. In: Life of freshwaters of the USSR, Vol. 4, Moscow, Leningrad, Russia: USSR Academy of Sciences, 183-265. (in Russian)
15. Klymiuk V., Barinova S. and Lyalyuk N., 2014 – Diversity and ecology of algal communities from the regional landscape park “Slavyansky Resort”, Ukraine, *Research and Reviews: Journal of Botanical Science,* 3, 2, 9-26.
16. Krupa E., Slyvinskiy G. and Barinova S., 2014 – The effect of climatic factors on the long-term dynamics of aquatic ecosystem of the Balkhash Lake (Kazakhstan. Central Asia), *Advanced Studies in Biology,* 6, 3, 115-136.
17. Krupa E. G., 2012 – Zooplankton of limnetic and running water ecosystems in Kazakhstan. Structure, forming patterns, Saarbrücken: Palmarium Academic Publishing, 346. (in Russian)
18. Kudekov T. K. (Ed.), 2002 – The modern ecological state of the basin of Lake Balkhash, Almaty, Kazakhstan: Kaganat, 386. (in Russian)
19. Mazurov A. K., 2005 – Metallogenic zoning of Kazakhstan, *Proceedings of Tomsk Polytechnic University,* 308, 4, 33-39. (in Russian)
20. Mingli Y., Cuixia Z. H., Zengjie J., Shujin G. and Jun S., 2014 – Seasonal Variations in phytoplankton Community Structure in the Sanggou, Ailian, and Lidao Bays, J. Ocean Univ. China (Oceanic and Coastal Sea Research), 13, 6, 1012-1024.

21. Moshkova N. A. and Gollerbach M. M., 1986 – Key to freshwater algae USSR, Vol. 10, 1, Green algae, Class Ulothrichophyceae, Moscow, Russia: Soviet Science, 361. (in Russian)
22. Muylaert K., Declerck S., Van Wichelen J., De Meester L. and Vyverma W., 2006 – An evaluation of the role of daphnids in controlling phytoplankton biomass in clear water versus turbid shallow lakes, *Limnologica*, 36, 69-78.
23. Nikanorov A. M. and Zhulidov A. V., 1991 – Biomonitoring of metals in freshwater ecosystems, Leningrad, Russia: Gidrometeoizdat, 144. (in Russian)
24. Novakovsky A. B., 2004 – Abilities and base principles of program module “GRAPHS”, *Scientific Reports of Komi Scientific Center, Ural Division of the Russian Academy of Sciences*, 27, 1-28.
25. Palamar–Mordvintseva G. M., 1982 – Key to freshwater algae USSR, Vol. 11, 2, Green algae. Class Conjugatophyceae. Desmidiaceae (2). Moscow, Russia: Soviet Science, 621. (in Russian)
26. Pieczy´nska E., Kołodziejczyk A. and Rybak J. I., 1999 – The responses of littoral invertebrates to eutrophication-linked changes in plant communities, *Hydrobiologia*, 39, 9-21.
27. Ponomareva L. P., Shaukharbaeva D. S. and Lopareva T. Y., 2005 – Hydrochemical variables and phytoplankton as trophy indicators of the lake Balkhash and Ily River’s reservoirs, In: Fisheries research in the Republic of Kazakhstan, Almaty, Kazakhstan: Kaganat, 366-376.
28. Popova T. G., 1955 – Key to freshwater algae USSR, Vol. 7, Euglenophyta, Moscow, Russia: Soviet Science, 213. (in Russian)
29. Samakova A. B. (Ed.), 2003 – Problems of hydroecological stability in the basin of Lake Balkhash, Almaty, Kazakhstan: Kaganat, 584.
30. Semenova A. D. (Ed.), 1977 – Guideline for chemical analysis of surface water. Leningrad, Russia: Gidrometeoizdat, 541. (in Russian)
31. Tarasov M. N., 1961 – Hydrochemistry of Balkhash Lake, Moscow, Russia: Academy of Sciences of the USSR, 233. (in Russian)
32. Tolotti M., Manca M., Angeli N., Morabito G., Thaler B., Rott E. and Stuchlik E., 2012 – Temperature modulated effects of nutrients on phytoplankton changes in a mountain lake, *Hydrobiologia*, 698, 61-75.
33. Vorobyeva N. B., Tyutenkov S. K., Sadukasova R. E. and Fokina A. S., 1982 – The current state and prospects for the development of the fodder base for fish in the Balkhash Lake depending on the level regime, In: Forecast of the integrated and rational use of natural resources. their protection and prospects for the development of the productive forces of the Lake Basin, Balkhash in the period until 1990-2000, Part 2, Alma-Ata, Kazakhstan: Science of KazSSR, 129-132. (in Russian)
34. Zabelina M. M., Kiselev I. A., Proshkina–Lavrenko A. I. and Sheshukova V. S., 1951 – Key to freshwater algae USSR, Vol. 4, Diatoms, Moscow, Russia: Soviet Science, 622. (in Russian)

THE ROLE OF DIASPORE BANKS FOR THE RESTORATION OF FLOODPLAIN MEADOWS. RESULTS OF A LONG TERM MONITORING ON THE NORTHERN UPPER RHINE (GERMANY)

Erika SCHNEIDER-BINDER *

* Karlsruhe Institute for Technology – University of Land Baden-Württemberg and Research Center of the Helmholtz Society, Institute for Geography and Geoecology, Department WWF-Institute for Floodplain Ecology, Josefstr. 1, Rastatt/Germany, D-76437, erika.schneider@kit.partner.edu; erika.schb@t-online.de

DOI: 10.1515/trser-2017-0010

KEYWORDS: diaspores in the soil, seed dispersal, seed longevity, succession stages of floodplain meadows.

ABSTRACT

The reconnection of the inner area of the island Kühkopf/Northern Upper Rhine to the hydrological dynamic of the river was the beginning of a large scale restoration programme for turning back to the previous vegetation – floodplain meadows. From the various aspects of the restoration are presented the succession phases; species with their different type of dispersal are analysed as well. A special attention is given to the role of diaspores of the soil, which play an important role in the redevelopment of floodplain meadows. Species of the diaspore bank can recur after many years, if favourable conditions are available. These are related to floods, dryness and rooting up by wild boar with the development of micro-succession stages.

ZUSAMMENFASSUNG: Die Rolle der Diasporenbank für die Renaturierung von Auenwiesen. Ergebnisse eines Langzeitmonitorings am nördlichen Oberrhein (Deutchland).

Die Wiederanbindung der inneren Bereiche des Kühkopfs an die Dynamik des Rheins war der Beginn eines umfassenden Programmes zur Wiederherstellung von Auenwiesen. Aus der Fülle der untersuchten Aspekte werden die unterschiedlichen Entwicklungsphasen vorgestellt und die Arten nach ihrem Verbreitungsmodus analysiert. Besondere Aufmerksamkeit gilt dem Diasporenreservoir des Bodens, das unter bestimmten Bedingungen nach Hochwasserereignissen, sommerlicher Trockenheit oder Brechen der Grasnarbe durch Wildschweine aktiviert wird. Dabei entstehen charakteristische Mikrosukzessionen.

REZUMAT: Rolul băncii de diaspori în redezvoltarea fâneţelor aluviale. Un monitoring de lungă durată în lunca sectorului nordic al Rinului superior (Germania).

Reconectarea părţii interioare a insulei Kühkopf la dinamica hidrologică a Rinului a constituit începutul unui larg program de reconstrucţie ecologică a fâneţelor de luncă, existente în zonă înaintea practicării unei agriculturi intensive. Dintre variatele aspecte ale refacerii pajiştilor de luncă sunt prezentate diferitele stadii de dezvoltare, fiind apoi analizate speciile cu modalitățile lor de dispersie. O atenţie specială e dată rolului diasporilor din sol. Speciile pot să apară după timp îndelungat, în caz că se ivesc condiţii favorabile de germinare. Acestea sunt legate de viituri, secetă estivală şi de activitatea de râmat a porcilor mistreţi, creându-se micro-succesiuni caracteristice.

INTRODUCTION

For the restoration of previously existing vegetation after change of land use, i.e. the transformation of agricultural lands into extensively used riverine floodplain meadows, dispersal units of plants are of great importance, they constitute a functional unit influencing the dynamics of populations and plant communities and should be considered in conservation and restoration programmes (Bornkamm et al., 1991; Poschlod, 1991; Curtean-Bănăduc et al., 2014). In particular the diaspore bank of the soil – seeds and fragments of rhizomes – plays an important role for restoration of floodplain meadows (Poschlod 1991, Dister et al. 1992).

The opportunity for floodplain restoration through natural succession (without the influence of man) and controlled by mowing arose on the Northern Upper Rhine in the floodplain area of Kühkopf, part of the Nature Reserve and Natura 2000 site "Kühkopf-Knoblochsaue" (Fig. 1).

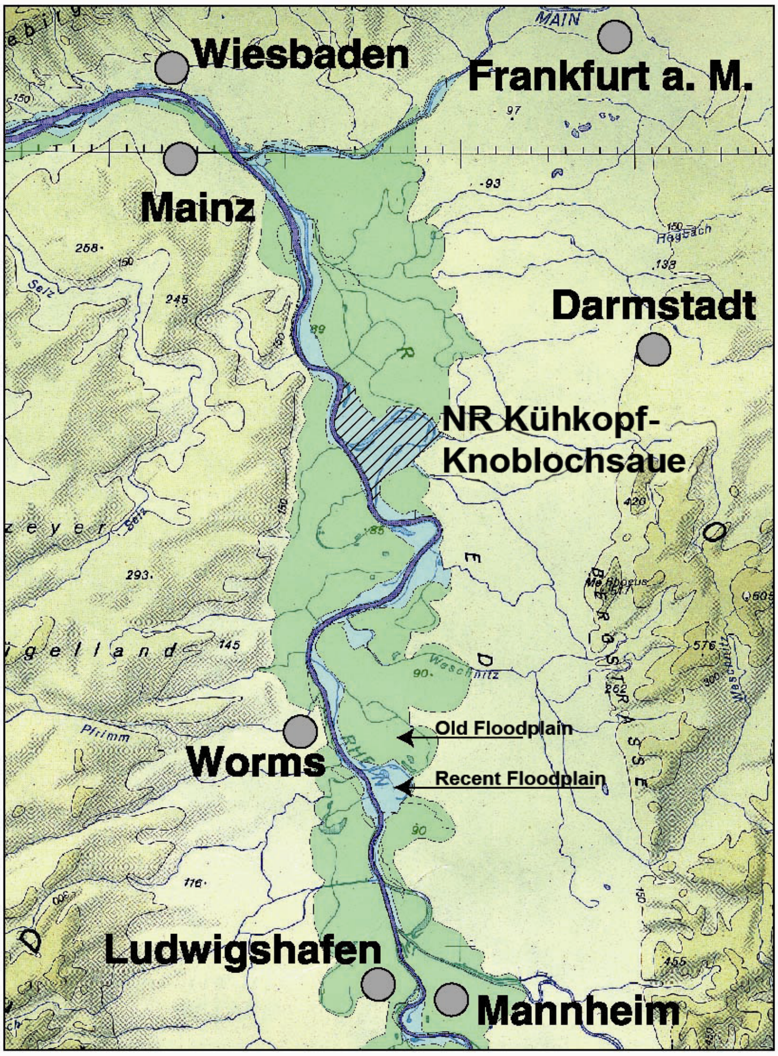


Figure 1: Geographical position of the Kühkopf on the Northern Upper Rhine (source: Auen-Institut, Rastatt).

The Kühkopf is an island that emerged from the cutting of a great meander of the river Rhine in 1828/1829. From that time the island has been surrounded by the Stockstadt-Erfelder Old Rhine and in its Western part by the New Rhine. The inner part of the island has been protected against floods by a system of dykes. In the three independent polders (A, B, C) with a total of 700 ha, on 400 ha agriculture – more recently intensively – has been practiced; 300 ha are covered by hardwood floodplain forest. The site's intensive use for crop breeding had constituted a permanent conflict between agriculture and nature protection. Then, after a dyke break as a consequence of high floods in spring 1983 with two peaks, the one in April and the next at the end of May, the agricultural use of the crop breeding station came to an end. The reconnection of the inner part of the area to the natural hydrological regime of the river Rhine and the decision to take the land out the agricultural use was the beginning of a large scale restoration programme with possibilities for a long-term monitoring on experimental plots scientifically followed from 1986 to 2001, 2005-2008 and 2014 and will be continued in subsequent years. (Dister et al.,1986; Dister et al., 1992; Schneider, 1995, 2001; Schneider et al., 1994)

The restoration raises many questions related to the phases of succession and the influencing factors which in floodplains are more complex than in the area outside the floodplains. They are strongly related to the flooding, the changes between high and low water levels, mainly the seasonal occurrence of floods, their height, duration and frequency, dry periods of the years and as well to the existing dispersal units of plants, the means of distribution and the competition capacity of the species and their populations. From the comprehensive volume of data monitored over more than two decades, this paper will present only some aspects related to the phases of the controlled succession by mowing and the role of the different dispersion of species with regard to the role of the diaspore bank of the soils.

The area subjected to natural and controlled succession in the past to a minor degree has been analysed from the point of view of dispersal biology (Schmidt, 1981). Only from the last two decades of the 20th century this type of analysis entered more and more into the attention of succession researches. The dispersal analysis can suggest the way of colonisation of species from the surrounding flora contributing to the restoration of the previous vegetation. Beside species of the diaspores bank of the soil, which can germinate in favourable conditions, in the first colonisation phase anemochorous dispersal, by wind-dispersed species, play an important role, species of other types of dispersal being on the beginning of succession of minor importance (see also Schmidt, 1981). Later the zoochorous dispersal plays as well an important role.

MATERIAL AND METHODS

In order to improve scientific knowledge in the field of succession of vegetation for restoration of floodplain meadows, permanent plots for natural succession (without the influence of man) for the development of floodplain forests and controlled succession with regularly mowing for restoration of floodplain meadows have been established on sites with various soil conditions (different content of sand and different micro-relief and height).

So as to document also the influence of game (wild boar, deer) on the course of succession, fenced and non-fenced plots have been established for each experimental area. A varied network of experimental variants has thus been established (Fig. 2). For the monitoring of controlled succession four areas were chosen; two in Polder A "Vor dem Eichwald"/A1 (altitude 86.7 m above NN) and "Ochsenlache"/A2 (altitude 86.25 m a. NN), one in Polder B

„Schafweide“ (altitude 85.8 m a.NN) and one in Polder C „Plattenacker“ (altitude 86.5 m a. NN) (Dister et al., 1987) (Fig. 2). The fenced and non-fenced plots of 10x30 m, were subdivided into smaller plot units, each including two 5x5 m quadrats (a1 and a2 non-fenced, b1 and b2 fenced), so that a total of sixteen identical permanent plots were included in the monitoring programme. As well a smaller area (1x1 m) for detailed studies of species distribution has been established, all plots being surrounded by a buffer zone. For the study of micro-successions the sampling area has been adapted to the extent of the area rooted up by wild boar. The quantitative and qualitative changes of the controlled succession for restoration of floodplain meadows on the abandoned agricultural lands were realized through comparative analysis of species composition, the distribution of the dominant species in the plot, the changes from one year to the other of the abundance and dominance of species and as well the change of structure and plant composition in the plot under the impact of different influencing factors such are high floods, soil rooting up by wild boar and summer dryness.

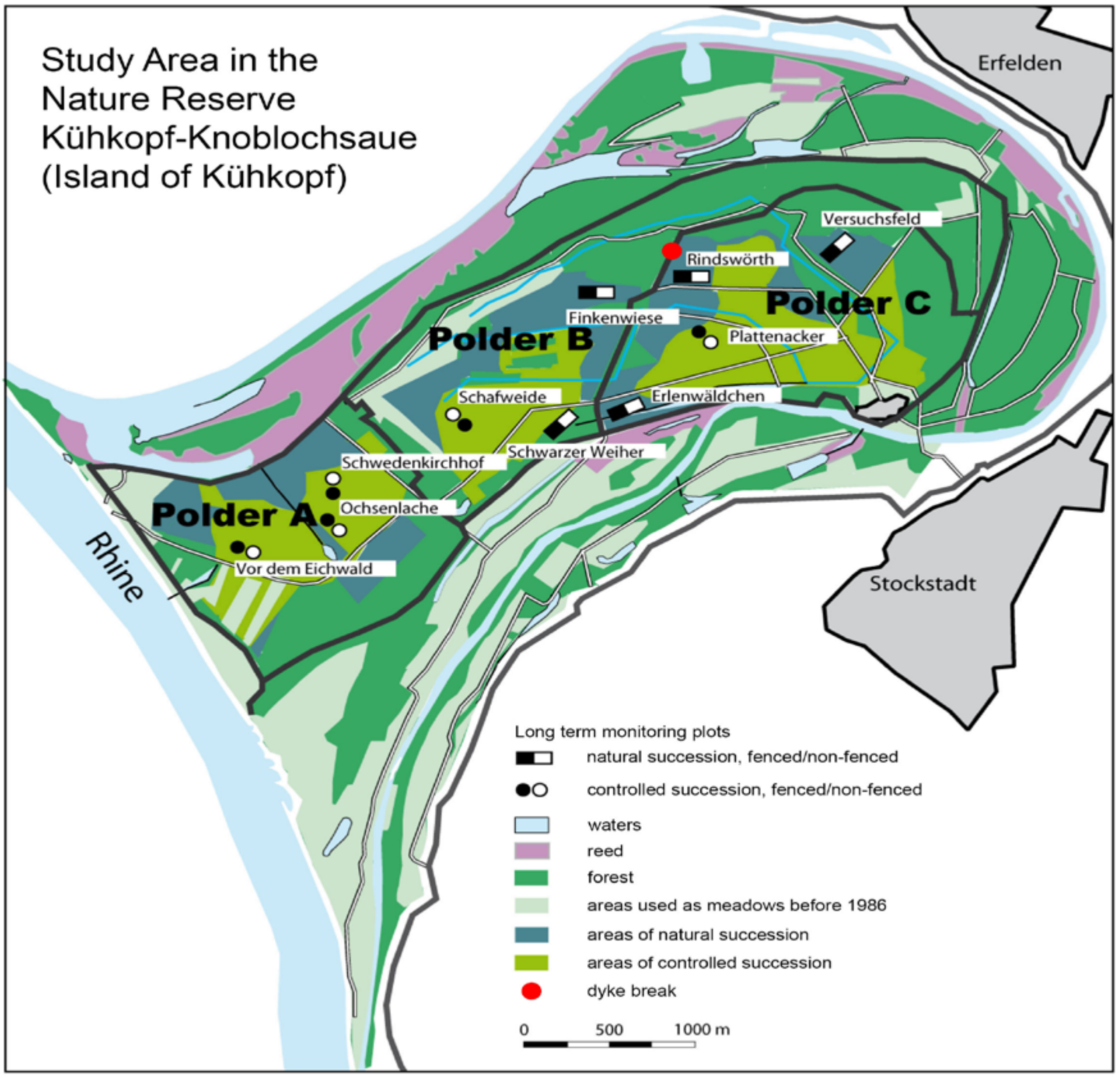


Figure 2: Study area on the Kühkopf, part of the Nature Reserve and Natura 2000 site Kühkopf-Knoblochsaue Northern Upper Rhine/Germany.

The plots were analysed annually by phytocoenological methods, according to Braun-Blanquet (1964), and as well other refined methods for the analysis of structure and sociability characteristics (Schmidt, 1981; Fischer, 1985; Dister et al., 1986, 1987; Schneider et al., 1994). The samples are included in phyto-coenological tables indicating for each species data about bioform, phytocoenological units, modality of dispersion and indicator value for wetness and nitrogen (Müller-Schneider, 1977) for dispersal units (Ellenberg et al., 2001).

The studied plots, fenced and non-fenced, were subjected to regular mowing, each year in the same time period from the beginning to the middle of June (depending on the flood events), harvested and removed from the area. Only in the first two years did mulching take place, the vegetation being open and the degree of cover very low. Depending on the weather in late August a second mowing took place.

RESULTS AND DISCUSSION

After the high floods of May 1983 and the definitive abandonment of agricultural use, colonization started in controlled plots (moving once or twice a year) and the succession went through various stages, from the stage of annual weeds of root crop (Chenopodietea) and cereal cultivation (Secalietea), i. e. Therophytes (in the first and the second year of succession 1983, 1984) to that of ruderal Hemicryptophytes (in the third, fourth and fifth year) and eventually the grass stage with herbs beginning in the fifth and sixth year (Dister et al., 1986 1987, 1990; Schneider, 2001, 2002).

Dominant species have been in the first stage, among others, *Chenopodium album, Polygonum persicaria* (*Persicaria maculosa*), *Amaranthus retroflexus, Amaranthius hybridus, Matricaria discoidea, Sonchus asper Coronopus squamatus, Conyza canadensis* and *Capsella bursa-pastoris* (Dister et al., 1992). Remarkable has been the occurrence on a large area of the solanaceous weeds *Hyoscyamus niger* and *Datura stramonium* (Polder A and C) and common poppy (*Papaver rhoeas*) (Polder B).

Due to the inner area of Kühkopf isolation by the surrounding Stockstadt-Erfelden old meander and a forest belt we have to assume that these species developed from the re-activated diaspore bank of the soil. The re-activation includes species from older agricultural land use and from former old floodplain meadows (Dister et al., 1992; Schneider 1995; Fischer, 1987).

In the third year (1985) the above-mentioned weeds mostly disappeared or occurred in a smaller area, a characteristic being at this stage a competition of annual weeds − all Therophytes − with also short-lived, mostly biennial Hemicrypothytes, indicating an early stage of ruderal tall herbaceous vegetation. The following stage of ruderal hemicryptophytes and geophytes with rhizomes, was dominated by creeping thistle (*Cirsum arvense*) and locally by couch-grass (*Elymus repens*) in the third and fourth year (partly in 1985 and in 1986) (Fig. 3). From the beginning of the succession the couch-grass became dominant in certain area.

As a consequence of the 1987 long summer flood and the selection between flood resistant, flood tolerant and flood intolerant species the thistle has been eliminated more rapidly, and common dandelion (*Taraxacum officinale*) has extended on some areas but in others has retreated. At the same time, stoloniferous grasses such as creeping bent (*Agrostis stolonifera*) and rough meadow-grass (*Poa trivialis*) have spread. This was the beginning of the establishing of a grass stage with herbs, in particular species from the pea family (Fabaceae), which developed micro-facies structures showing “flowerbed” aspects (Fig. 4). This structure remained for 4-5 years and changed more or less rapidly due to fluctuating water tables. Finally, regular cutting altered these micro-facies structures and the area’s aspect became similar to natural floodplain meadows dominated by different grass stages.

In the eighth year and even more in the ninth year, the grasslands growing in the more elevated places turned into Arrhenatheretum elatioris-like meadows, but still a typical composition and many of the characteristic were lacking. Adjacent to the tall oat-grass (*Arrhenatherum elatius*) meadows, but in the lower-lying spots, the floodplain meadows were dominated by meadow foxtail (*Alopecurus pratensis*), this last having a higher wetness tolerance than tall oat grass (Ellenberg et al., 2001). In the lowest places, i.e. the more or less regularly flooded sites, one finds wet meadows with swamp meadow-grass (*Poa palustris*). In the further course of succession, in the twelfth year of meadows development (1994) tall oat-grass (*Arrhenatherum elatius*), meadow foxtail (*Alopecrus pratensis*) and swamp meadow-grass (*Poa palustris*) covered larger area according to their wetness requirements, the swamp meadow-grass being restricted to the lowest levels, the existing small depressions and flood channels. With the development and spreading of the tall oat grass and the meadow foxtail, characteristic accompanying species for these communities appeared, such as *Galium mollugo*, *Centaurea jacea* and *Campanula patula*. In the foxtail-meadows *Inula salicina* and *Lychnis flos-cuculi* occurred on some places (Schneider, 2002).

Figure 3: Stage of ruderal Hemicrypthophytes, dominated by Creeping thistle (*Cisium arvense*) with smaller wet-tolerant grasses on the low level.

Figure 4: Micro-facies structures showing “flowerbed” aspects with red clover (*Trifolium pratense*) and ox-eye daisy (*Chrysanthemum leucanthemum*).

Figure 5: Grass stage in the sampling area with dominant tall fescue (*Festuca arundinacea*) and tall oat-grass (*Arrhenatherum elatius*).

As a consequence of the dry summer of 1991 a large disturbance of the plots took place, many species susceptible to dryness such as *Poa palustris*, *P. trivialis*, *Potentilla reptans* and others disappeared (Dister et al., 1991) and occurred again two years later. After the summer flood of 1999 large changes in the abundance of different species toke place in the grasslands cover of the Kühkopf area. The degree of cover of *Arrhenatherum elatius* and *Alopecurus pratensis* decreased and tall fescue (*Festuca arundinacea*) spread and dominated over a large area, a situation during more years and existing as well today in large parts of the polders, including the monitored plots.

The development in relation to the hydrological regime, the changes between flooding and drying out, are the influencing factors as well for the re-activation of diaspores of the soil. They play an important role not only in the first colonization of the abandoned agricultural lands, but also in the course of succession, when disturbances such are, high floods, dryness and rooting up by wild boars arise. Dry summertimes combined with rooting up by wild boar are the basis for the re-activation and spreading of xero-thermophilous species from the diaspore reservoir of the soil.

Near the species from the diaspore bank of the soil, occurring if site conditions are good, anemochorous wind-dispersed species play a main role in the first colonization phase (Schmidt, 1981), and in the later succession course if changing ecological conditions. From the species with other dispersal mechanisms the zoochorous and anemo-zoochorous species play an important role (Fig. 6 and 7). The species number as a whole and the number of anemochorous, zoochorous and anemo-zoochorous species is variable from year to year in function of the flood events and dryness in the area. Remarkable changes over the years are exemplified by two sampling data of the year 1987, before and after a high flood with long duration with remarkable differences in species number as a whole and as well from the three categories of seed dispersion.

The activation of the diaspore bank includes different aspects. As in the studied case, diaspores with long-term persistence occurred – mostly weeds from former traditional agricultural land use and housing, as well diaspores of species from older floodplain meadows such as *Viola elatior*, *Anacamptis pyramidalis* and *Ranunculus bulbosus*, which existed before the agricultural land use and survived as diaspores in the soil. As it is stated during other researches diaspores are concentrated on the upper 30 cm of the floodplain soil (around 60-80% of the diaspores). Diaspores can rarely be found deeper than 50 cm (Hölzel and Otte, 2003). But a special case of persistence of diaspores in the seed bank of soils is given by the black henbane (*Hyoscyamus niger*) in the succession area of Kühkopf. The species colonized large area on the beginning of the succession in the first and second year (after dam breaking), disappearing again for a time. In 1992 it occurred again in places rooted up by wild boar. These rooted up area constitute the beginning of a new micro-succession in between the larger area of succession in a more advanced phase of evolution to meadows (Schneider, 2002). Later occurrence has been registered in 1998 and 2005 also in the wild boar rooting up area. The occurrence of the black henbane, a species with long-term persistence in the diaspore bank of the soil was reported also in other parts of the Upper Rhine, after large and deep earth moving for hydro-technical construction (oral communication by Prof. Dr. Roland Carbiener, Strasbourg, France). From these observations it can be stated that the seeds of black henbane can survive also in deeper soil levels and from older times. The longevity of the seeds of black henbane has been mentioned up to 650 years (Poschlod, 1991, according to Odum, 1965).

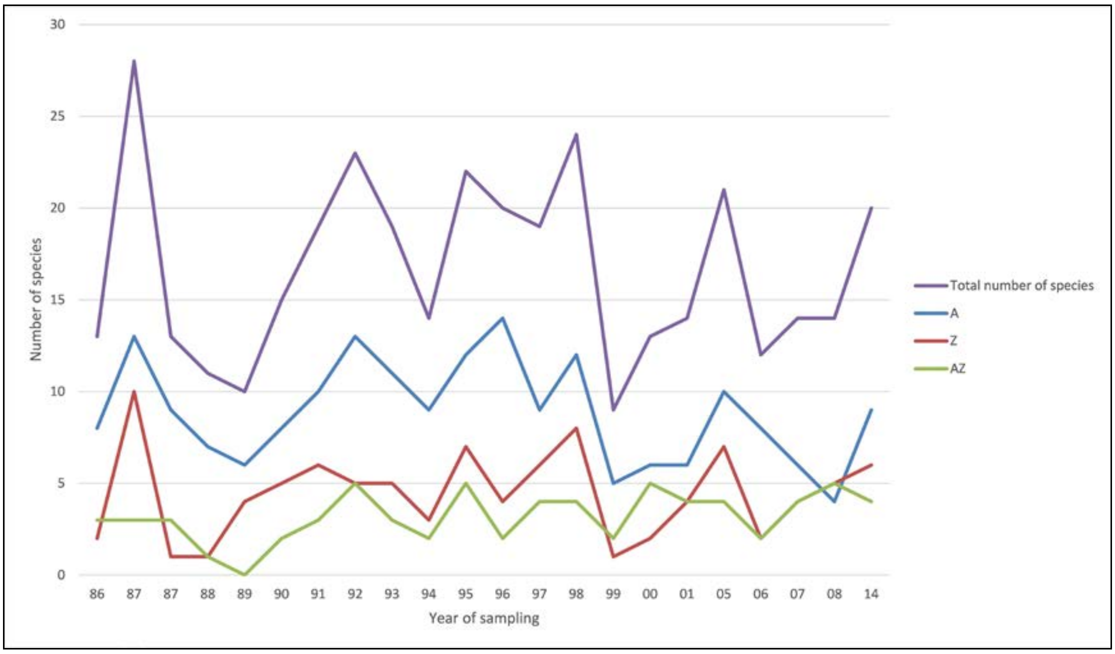


Figure 6: Number of anemochorous (A), zoochorous (Z) and anemo-zoochorous species in comparison with the total number of species in the sampling plots A2 "Ochsenlache" from 1986-2001, 2005-2008 and 2014; the year 1987 is represented with two samples before and after a high flood of long duration.

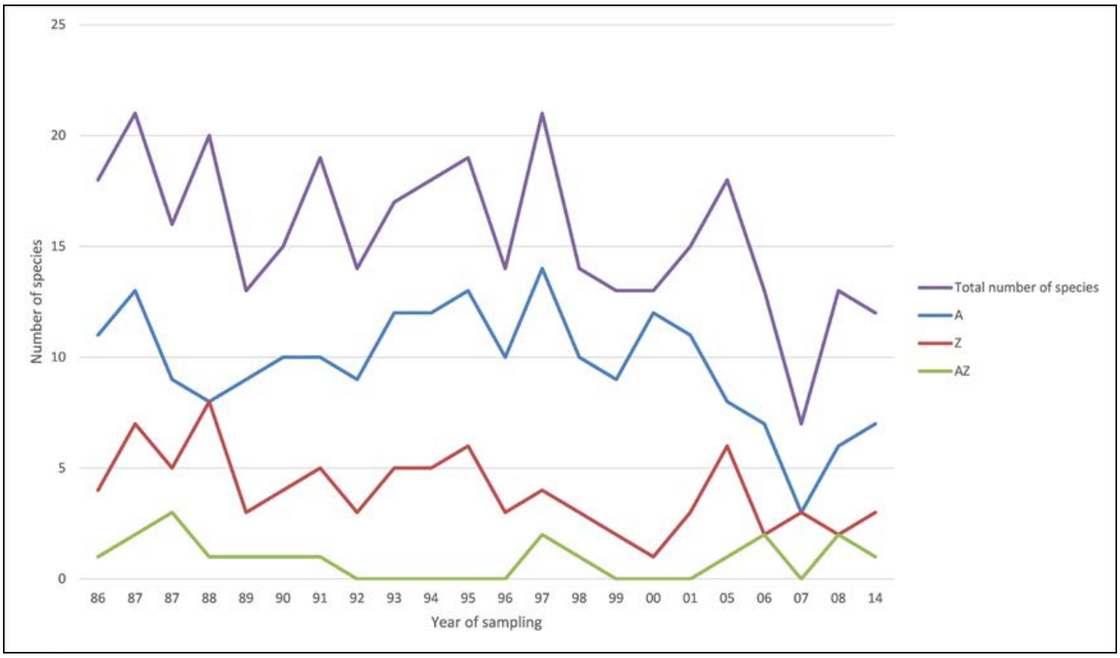


Figure 7: Number of anemochorous (A), zoochorous (Z) and anemo-zoochorous (AZ) species in comparison with the total number of species in the sampling plots of the area C "Plattenacker" from 1986-2001, 2005-2008 and 2014.

Figure 8: Micro-succession in a meadow-like succession stage with species occurred from the reactivation of the seed bank of the soil (1992).

On wild boar rooting up sites there occurred in 1993 a typical community of weeds of old agricultural lands, Mercurialetum annuae Krusem. Et Vlieg. 39 em. Th. Müller, composed of the thermophilous, sub-Mediterranean, sub-Atlantic species *Mercurialis annua* and including a large number of other thermophilous species such as *Datura stramonium*, *Hyoscyamus niger*, *Euphorbia helioscopia*, *Lathyrus tuberosus*, and others (Tab. 1). This plant community, indicating old land use by root crop cultures (Oberdorfer, 1983), has to be considered also as a reactivation of the diaspore bank of the soil. This community became very rare on the Upper Rhine and in the last decades up to the 1970s has not been observed, being considered as having disappeared. During the studies and long term monitoring the community has been observed in 1993 (Schneider and Dister, 1993; Schneider, 1995) and also later in 1998 and 2005 (unpublished data).

Table 1: Mercurialetum annuae Krusem. et Vlieg. 39 em. Th. Müller facies with *Datura stramonium* and *Hyoscyamus niger*.

Number of sample			1	2	3	4	5	6	7	8	9	10	
Size of sample			25	20	25	35	12	20	113	30	30	12	
Coverage degree %			65	50	30	40	45	75	60	60	30	55	
Biof.	PhSoc												C
T	FE	*Veronica polita*	+	+	.	+	+	+	.	.	.	+	III
T	FE	*Mercurialis annua*	+	1	+	.	+	.	+	+	1	2	V
T	FE	*Thlaspi arvense*	.	.	.	.	.	+	.	.	.	+	I
T	PCH	*Chenopodium polyspermum*	1	+	+	+	2	3	1	2	+	2	V
T-H	PCH	*Lamium purpureum*	.	.	.	+	+	+	.	+	.	.	II
T	PCH	*Lamium amplexicaule*	.	.	.	+	+	.	.	.	+	.	II
T	PCH	*Polygonum persicaria*	.	.	.	.	+	.	.	+	+	+	II
T	PCH	*Euphorbia helioscopia*	.	.	.	.	+	.	.	.	.	.	I
T	CH	*Datura stramonium*	3	3	2	3	+	.	+	1	2	2	V
T	CH	*Chenopodium album*	.	.	+	+	+	1	1	.	+	+	IV
T	CH	*Chenopodium hybridum*	.	.	.	+	+	1	3	2	+	.	III
T	CH	*Chaenorhinum minus*	.	.	.	.	.	.	+	.	.	.	I
T	CH	*Atriplex patula*	.	.	.	.	.	.	+	.	+	.	I
T	CH	*Solanum nigrum*	+	1	+	2	1	2	.	+	+	+	V
T	CH	*Amaranthus retroflexus*	.	.	+	.	+	+	+	+	.	.	III
G	CH	*Sonchus arvensis*	.	+	+	+	.	.	.	.	.	.	II
T	CH	*Capsella bursa pastoris*	+	.	+	+	.	+	.	.	.	+	III
T	SI	*Sisymbrium officinale*	+	.	.	.	.	.	.	.	.	.	I
T	SI	*Brassica napus*	.	.	+	.	.	.	.	.	.	.	I

Table 1 (continued): Mercurialetum annuae Krusem. et Vlieg. 39 em. Th. Müller facies with *Datura stramonium* and *Hyoscyamus niger*.

Number of sample			1	2	3	4	5	6	7	8	9	10	
Size of sample			25	20	25	35	12	20	11 3	30	30	12	
Coverage degree %			65	50	30	40	45	75	60	60	30	55	
Biof.	PhSoc												C
T	CAU	*Euphorbia exigua*	.	.	.	.	.	.	+	.	.	.	I
G-H	CAU	*Lathyrus tuberosus*	.	+	+	+	+	+	+	+	+	+	V
T	SE	*Viola arvensis*	.	.	+	+	.	.	+	.	.	.	II
T	SE	*Anagallis arvensis*	+	.	.	.	+	.	.	.	.	.	I
T	SE	*Papaver rhoeas*	.	.	.	+	.	.	.	.	.	.	I
G	SE	*Convolvulus arvensis*	+	+	.	.	+	+	.	.	.	+	III
T-H	ON	*Hyoscyamus niger*	.	.	.	+	1	1	1	2	.	.	III
H	ON	*Carduus acanthoides*	.	.	.	+	.	.	+	.	.	.	I
H	ART	*Cirsium vulgare*	.	.	.	.	.	.	+	.	.	.	I
H	ART	*Dipsacus laciniatus*	.	.	.	.	.	.	+	.	.	.	I
H	ART	*Daucus carota*	.	+	.	.	.	.	.	.	.	.	I
G	ART	*Cirsium arvense*	+	+	+	.	.	2	.	.	.	1	III
T	ART	*Galium aparine*	.	.	.	.	.	.	+	.	.	.	I
H	AR	*Arrhenatherum elatius*	.	.	.	+	+	+	.	.	.	.	II
C-H	AR	*Trifolium repens*	.	.	.	.	.	+	.	.	.	+	I
H	AR	*Lolium perenne*	.	.	.	.	.	.	.		+	+	I
H	AR	*Chrysanth. leucanthemum*	.	.	.	.	.	.	.	.	+	.	I
H	MA	*Trifolium pratense*	.	.	+	.	.	.	.	.	.	.	I
H	MA	*Dactylis glomerata*	.	.	.	1	2	.	+	.	.	.	II

Table 1 (continued): Mercurialetum annuae Krusem. et Vlieg. 39 em. Th. Müller facies with *Datura stramonium* and *Hyoscyamus niger*.

Number of sample			1	2	3	4	5	6	7	8	9	10	
Size of sample			25	20	25	35	12	20	11 3	30	30	12	
Coverage degree %			65	50	30	40	45	75	60	60	30	55	
Biof.	PhSoc												C
H	MA	Taraxacum officinale	1	1	1	+	2	1	+	+	2	+	V
G	AgR	*Elymus repens*	2	1	1	+	.	.	+	.	.	.	III
G-H	AgR	*Rorippa sylvestris*	.	.	.	+	.	.	.	.	.	.	I
H	AgR	*Festuca arundinacea*	.	.	.	.	.	.	+	.	.	.	I
H	AG	*Potentilla reptans*	.	.	.	.	.	.	2	+	.	.	I
T	PL	*Polygonum aviculare*	+	.	+	+	+	.	+	+	+	+	IV
H	PL	*Plantago major*	.	.	.	+	+	+	.	.	.	.	II
T-H	ME	*Medicago lupulina*	.	.	.	+	.	+	.	.	.	.	I
H	ME	*Euphorbia esula*	.	.	.	.	.	.	.	+	.	.	I
H	TR	*Agrimonia eupatoria*	.	.	.	.	+	+	.	+	.	.	II
H	PH	*Poa palustris*	.	.	.	.	.	+	.	.	.	.	I
P	AU	*Pyrus pyraster*	.	.	.	.	.	.	.	+	.	.	I
C	CO	*Rubus caesius*	.	.	.	.	.	.	.	+	.	.	I

Abbreviations of the phyto-coenological units used in the table: FE = Alliance Fumario-Euphorbion; PCH = Ordre Polygono-Chenopodietalia; CH = Classe Chenpodietea; SI = Alliance Sysimbrion officinals; CAU = Alliance Caucalidion lappulae; SE = Ordre Seacalietalia and Classe Secalietea; ON = Ordre Onopordetalia; ART = Classe Artemisietea; AR = Ordre Arrhenatheretalia; MA = Classe Molinio-Arrhenatheretea; AgR = Alliance Agropyro-Rumicion; AG = Classe Agrostietea stoloniferae; PL= Classe Plantaginetea; ME = Alliance Mesobromion erecti; TR = Alliance Trifolion medii; PH = Classe Phragmitetea; AU = Alliance Alno-Ulmion; CO = Ordre Convolvuletalia.
Date of sampling: Sample 1-9, 9.9.1992, sample 10 16.9.1992.

In years with longtime floods, in the deeper flood channels, i.e. depressions of the newly developing meadows, occur sometimes short-lived annual species adapted to wetness. They colonise the meadow type of *Poa palustris*, building open pioneer communities. These communities are dominated by *Potentilla supina* accompanied by other short-lived species such *Gnaphalium luteo-album, Chenopodium* spp. and *Polygonum* spp., all being reactivated from the diaspore bank of the soil. Some species have a high longevity in the soil, i.e. *Fumaria officinalis*, being mentioned more than 500 years (Oberdorfer, 2001). *Mercurialis annua*, *Potentilla supina* and *Chenopodium botryoides,* as well as *Chenopodium polyspermum,* occur only from time to time, as a function of the ecological condition of the year.

Table 2: *Potentilla supina* – community of short living species.

	Number of sample	1	2	3	4	5	6
	Coverage degree %	20	70	60	60	55	45
Biof.							
T(H)	***Potentilla supina***	**1**	**3**	**1**	**3**	**2**	**1**
T	*Capsella bursa pastoris*	1	+	+	+	+	+
T	*Polygonum persicaria*	.	+	.	+	.	.
T	Chenopodium album	+	.	+	.	.	.
T	*Chenopodium polyspermum*	1	.	.	.	.	.
T	*Chenopodium viride*	.	.	.	+	+	.
T	*Chenopodium botryoides*	.	2	.	.	.	.
T	*Gnaphalium luteoalbum*	.	**2**	.	.	.	.
T	*Polygonum lapathifolium*	.	+	.	.	.	.
T	*Anagallis arvensis*	.		1	+	+	+
T	*Lamium amplexicaule*	.	.	+	+	+	+
T	*Thlaspi arvense*	.	.	.	+	.	.
T	*Polygonum aviculare*	.	.	+	.	.	.
T	*Atriplex hastata*	.	.	.	+	+	.
T	*Fumaria officinalis*	.	.	.	.	.	+
T	*Euphorbia helioscopia*	.	.	.	.		+
H	*Taraxacum officinale*	.	+	3	3	2	3
H	*Plantago major*	.		+	+	+	+
H	*Plantago intermedia*	.	+	.	.	.	.
H	*Trifolium repens*	.	.	1	+	.	.
H(T)	*Medicago lupulina*	.	+	.	+	+	.
H	*Rumex crispus*	.	.	+		+	.
H	*Erigeron annuus*	.	.	+	.	+	.
H	*Poa palustris*	.	.	+	.	.	.
H	*Cirsium vulgare*	.	.	+	+	.	.
H	*Vicia cracca*	.	.	+	.	+	.
H	*Alopecurus pratensis*	.	.	.	+	.	.
H	*Rumex obtusifolius*	.	.	.	.	+	.
G-H	*Rorippa sylvestris*	.	+	+	+	.2	+
G	*Agropyron repens*	+	.	.	.	.	.
G	*Cirsium arvense*	.	.	.	.	+	.

Sampling data and place: 1 = 13.06.1994 plot “Schafweide”, Polder B; 2= 5.08. 1999, in the flood channel of “Plattenacker”, Polder C; 3-6 = 8.06. 2000 “Plattenacker” flood channel.

CONCLUSIONS

It is quite obvious that in floodplains the succession process is different from the succession in wet fallow lands outside the floodplain. This difference is caused by the coincidence of the flooding factor with other ecological conditions.

The succession progresses very rapidly and follows different stages from the initial pioneer therophytes to different structured stages of herbs and grasses. At the same time the studies show that a considerable genetic potential with numerous rare and endangered species can be activated by significant and unknown residues and by the seed bank in the soil. This last plays an important role in the evolution of the area.

It became clear in the course of the monitoring, that there are many species re-colonising the area only through the mechanism of diaspores of the soil (see also Fischer, 1987; Poschlod, 1991).

The reactivation of the seed bank in the soil is in strong relation to the flooding regime of each of the studied years and the occurrences of wet and dry periods. Longer wet and dry periods create different conditions in the vegetation cover influencing the vitality of species in function of their flood tolerance or capacity to survive dry summers. The gaps arising in the turf favour the colonization by short-lived species from the seed bank of the soil.

Rooting up by wild boar creates as well open larger or smaller patches in the developing meadows, leading to the development of micro-succession, with many species present from the seed bank of the soil.

Diaspore banks influence the dynamics of these plant communities and should be considered in the programmes of sustainable development and conservation of floodplain meadows.

ACKNOWLEDGEMENTS

During the long time of research and monitoring in the frame of the succession project, we benefited from the technical support and help of the Forestry Office Forstamt Groß-Gerau /Kühkopf Knoblosaue. In particular I would like to express my gratitude to Mr. Baumgärtel R. for its valuable help, fruitful discussions and assistance during field activities in the study area of the Nature Reserve Kühkopf-Knoblohsaue.

REFERENCES

1. Bornkamm R., Eggert A., Küppers M., Schmid B. and Stöcklin J. (Editors), 1991 – Liste populatiosbiologisch relevanter Begriffe. In: Schmid B & Stöcklin J. - Populationsbiologie der Pflanzen. Birkhäuser Verlag Basel- Boston – Berlin, 9-13. (in German)
2. Braun-Blanquet J., 1964 – Pflanzensoziologie. Grunzüge der Vegetationskunde, Auflage, Springer Verlag Berlin, Wien, New York, 3, 865. (in German)
3. Curtean-Bănăduc A., Schneider-Binder E. And Bănăduc D., 2014 – The importance of the riverine ligneous vegetation for the Danube Basin lotic ecosystems, in Cianfaglione K. (ed.) L'importanza degli Alberi e del Bosco, Cultura, scienza e coscienza del territorio, Temi Ed., Trento, Italia, ISBN: 978-88-973772-63-9, I-II, 187-210.
4. Dister E., Schneider E., Schneider Eckb., Fritz H.-G., Flößer E. and Winkel S., 1986 – Erfassung der tierischen und pflanzlichen Sukzession auf den aufgelassenen Ackerflächen des Kühkopfs im NSG „Kühkopf-Knoblochsaue, im Auftrag des Landes Hessen vertreten durch den Hessischen Minister für Umwelt und Energie in Wiesbaden, 113. (in German)
5. Dister E., Schneider E., Schneider E., Fritz H.-G., Winkel S. and Flößer E.,1987 − Erfassung der tierischen und pflanzlichen Sukzession auf den aufgelassenen Ackerflächen des Kühkopfs im NSG "Kühkopf-Knoblochsaue". Forschungsergebnisse 1987, Bericht, WWF-Auen-Institut Rastatt, erarbeitet im Auftrag der BFN, Darmstadt, 168. (in German)
6. Dister E., Schneider E., Fritz H.-G., Flößer E., Schneider E. and Winkel S., 1991 − Erfassung der Sukzession auf den aufgelassenen Ackerflächen des Kühkopfs im NSG "Kühkopf-Knoblochsuae", Ergebnisse 1991, WWF-Auen-Institut, Rastatt, im Auftrag der BFN, Darmstadt, 203. (in German)
7. Dister E., Schneider E., Schneider E., Fritz H.-G., Winkel S. and Flößer E., 1992 − Großflächige Renaturierung des "Kühkopfes" in der hessischen Rheinaue - Ablauf, Ergebnisse und Folgerungen der Sukzessionsforschung, in: Auen - gefährdete Lebensadern Europas. Renaturierung von Flussauen. Tagungsdokumentation des internationalen Kongresses in Rastatt; Beiträge der Akademie für Natur- und Umweltschutz Baden-Württemberg, 13 b, 20-36. (in German)
8. Ellenberg H., Weber H. E., Düll R., Wirth V. and Werner W., 2001 – Zeigerwerte von Pflanzen in Mitteleuropa, *Scripta Geobotanica*, 18, 1-264, Göttingen. (in German)
9. Fischer A., 1987 – Untersuchungen zur Populationsdynamik am Beginn von Sekundärsukzessionen. Dissertationes Botanicae, 110, Berlin-Stuttgart, 234. (in German)
10. Hölzl N. und Otte A., 2001 − The impact of flood regime on the soil seed bank of flooded meadows, *Journal of Vegetation Science*, 12, 209-218.
11. Hölzl N. und Otte A., 2003 – Restoration of a species rich flood meadow by top soil removal and diaspore trabsfer with plant material, *Apllied Vegetation Science*, 6, 131-140.
12. Müller-Schneider P., 1977 – Verbreitungsbiologie (Diasporologie) der Blütenpflanzen.- Veröffentlichungen des Geobotanischen Institutes der ETH, Stiftung Rübel, Zürich, volume 61, 226. (in German)
13. Oberdorfer E., 1993 – Süddeutsche Pflanzengesellschaften, III, 455, Fischer Verlag Jena. (in German)
14. Oberdorfer E., 2001 − Pflanzensoziologische Exkursionsflora für Deutschland und angrenzende Gebiete, 8, Auflage,. Verlag Eugen Ulmer Stuttgart, 1051. (in German)
15. Poschlod P., 1991 − Diasporenbanken in Böden - Grundlagen und Bedeutung, in: Populationsbiologie der Pflanzen, hrsg. von B. Schmid & J. Stöcklin: 15-35, Birkhäuser, Basel-Boston Berlin. (in German)
16. Schmidt W., 1981 – Ungestörte und gelenkte Sukzession auf Brachäckern, *Scripta Geobotanica*, 15, 1-199, Göttingen. (in German)

17. Schneider E., 1995 − Zur Vegetationsentwicklung auf den aufgelassenen Ackerflächen des Kühkopfs und das damit verbundene Auftreten seltener Arten. Zeitschrift für Vogel, und Naturschutz in Südhessen, Collurio 13, 67-78, Arbeitskreis Darmstadt der Hessischen Gesellschaft für Ornithologie und Naturschutz e. V. (HGON). (in German)
18. Schneider E., 2001 – Restoration of floodplain meadows and forests. Results of 15 years of monitoring in natural and controlled succession on re-flooded areas in the Nature Reserve Kühkopf-Knoblochsaue/Upper Rhine, River restoration in Europe. Practical approaches. Conference on river restoration Wageningen, The Netherlands 2000, Proceedings edited by H. J. Nijland & M. J. R. Cals, RIZA rapport nr. : 2001.023, 197-199.
19. Schneider E., 2002 − Vom Acker zur Auenwiese. 20 Jahre Grünlandsukzession auf dem Kühkopf, in 50 Jahre Naturschutzgebiet Kühkopf-Knoblochsaue. Regierungspräsidium Darmstadt, 43-49. (in German)
20. Schneider E. and. Dister E., 1993 − Erfassung der Sukzession auf den aufgelassenen Ackerflächen des Kühkopfs im NSG "Kühkopf-Knoblochsaue". Ergebnisse 1993, WWF-Auen-Institut, Rastatt, im Auftrag des Hessischen Ministeriums für Landesentwicklung, Wohnen, Landwirtschaft, Forsten und Naturschutz, 8, Project report. (in German)
21. Schneider E., Dister E. and Schneider E., 1994 − Erfassung der Sukzession auf den aufgelassenen Ackerflächen des Kühkopfs im NSG "Kühkopf-Knoblochsaue" Ergebnisse, WWF-Auen-Institut, Rastatt, im Auftrag des Hessischen Ministeriums für Landesentwicklung, Wohnen, Landwirtschaft, Forsten und Naturschutz, 101, Project Report. (in German)

DISTRIBUTION AND ABUNDANCE OF *ARTEMIA SALINA* IN THE SALT LAKE BASIN (CENTRAL ANATOLIA, TURKEY)

Ali ALAŞ *, *Murat KAYA* *, and *Ahmet ÖKTENER* *

* Necmettin Erbakan University, Faculty of A. K. Education, Department of Biology Education, Meram-Konya, Turkey, TR-42090, alasali@hotmail.com
** Aksaray University, Faculty of Science and Letters, Department of Biology, Aksaray, Turkey, TR-68100, muratkaya3806@yahoo.com
*** Sheep Research Institute, Department of Fisheries, Çanakkale street 7 km., Bandırma, Balıkesir, Turkey, TR-10200, ahmetoktener@yahoo.com

DOI: 10.1515/trser-2017-0011

KEYWORDS: *Artemia salina*, distribution, under threat, Salt Lake, Turkey.

ABSTRACT

In this study, the distribution and abundance of *Artemia salina* in 10 different stations of the Salt Lake Basin were investigated. In addition, its relationship to pH, dissolved oxygen, temperature, electrical conductivity and water levels were analyzed. Field studies were carried out from July to August of 2010. *Artemia salina* was observed in five of these stations. *A. salina* was not seen in some stations that have high electrical conductivity. It is determined that, in the station named Tersakan Lake where electrical conductivity was 154 mS/cm, *A. salina* is more abundant when compared to the other stations. But as underground water pumps that are built for the irrigation of agricultural lands decrease water levels, *Artemia salina*'s life is under threat.

RESUMEN: Distribución y abundancia de *Artemia* en la Cuenca del Lago Salado (Central Anatolia, Turquia).

En este estudio, la distribución y la abundancia de *Artemia salina* en 10 diferentes estaciones de la cuenca del Lago Salado fueron investigadas. Además, su relación con el pH, oxígeno disuelto, temperatura, conductividad eléctrica y nivel del agua fueron analizados. Estudios de campo fueron llevados a cabo desde julio hasta agosto de 2010. La *Artemia salina* fue observada en cinco de las estaciones de estudio. La *Artemia salina* no fue vista en algunas de las estaciones que tienen alta conductividad eléctrica. Se determinó que, en la estación llamada Lago Tersakan, donde la conductividad electrica fue de 154 mS/cm, la *A. salina* es mas abundante cuando comparada con otras estaciones. Pero como las bombas de agua subterráneas que se construyen para el riego de las tierras agrícolas disminuyen el nivel del agua, la vida de *A.a salina* está bajo amenaza.

REZUMAT: Distribuția și abundența speciei *Artemia salina* în cuveta Lacului Tuz (Anatolia Centrală, Turcia).

În prezentul studiu, s-au analizat distribuția și abundența speciei *Artemia salina* în 10 puncte de prelevare diferite din lacul Tuz. De asemenea, au fost analizate efectele pH-ului, ale oxigenului dizolvat, temperaturii, conductivității și nivelului apei asupra celor doi parametri populaționali considerați. Cercetările de teren au fost efectuate în perioada iulie-august 2010. *Artemia salina* a fost observată în cinci din cele 10 stații. *Artemia salina* a lipsit în unele stații, în care apa prezintă cantități mari de săruri dizolvate (conductivitate electrică mare). S-a concluzionat că în stația Tersakan Lake unde conductivitatea a fost de 154 mS/cm, *Artemia salina* este mai abundentă decât în celelalte stații. Din pacate, forajele care pompează apă din pânză freatică pentru a fi folosită pentru irigarea terenurilor agricole scad nivelul apei, amenințând existența speciei.

INTRODUCTION

Artemia salina appear naturaly in salt lakes, is used as live bait in aquariums, freshwater and marine fish culture, and as the main diet of some crustacean species because of its high nutritional value (Bengtson et al., 1991; Lengyel et al., 2012).

Artemia salina, which constitutes the second level of the food chain in the Salt Lake, feeds on suspended particles, bacteria and algae.

Similarly, flamingos and some other birds feed on *Artemia salina*.

This is why *Artemia salina* has a big importance on the energy cycle in the Salt Lake.

According to Başbuğ (1999), the *Artemia salina* species living in the Salt Lake, and it reproduces parthenogenetically.

In another research on *Artemia salina* in 1995 (Başbuğ, 1999), reproduction aspects and fecundity were analyzed.

This study was carried out in order to determine and reveal the distribution of *Artemia salina* in the Salt Lake Basin, its abundance and its relationship with water parameters in 10 different stations.

MATERIAL AND METHODS

Water was taken a total of 8 times (200 liters) from each of the 10 stations (Tab. 1, Fig. 2) in the Salt Lake basin; the water was filtered with a 55 µm plankton net and put into 500 ml. plastic bottles.

Right after this, 4% formaldehyde was added into the filtered samples and they were preserved.

The samples were brought to the laboratory and counted under a stereo microscope; individual numbers in the 200 liters were calculated and individual number per 1 m^3 was determined.

On the other hand, some water parameters were calculated in this study.

Study area

Salt Lake (Fig. 1a, b) is situated at the center of Turkey (Fig. 2).

It is the second biggest lake in terms of square measure; it has borders with Aksaray, Konya and Ankara cities.

More than half of the country's salt need is supplied by this lake. Surface area of the lake is 1500 km^2 and the altitude is 905 meters.

Figure 1a: Salt Lake.

Figure 1b: Salt Lake.

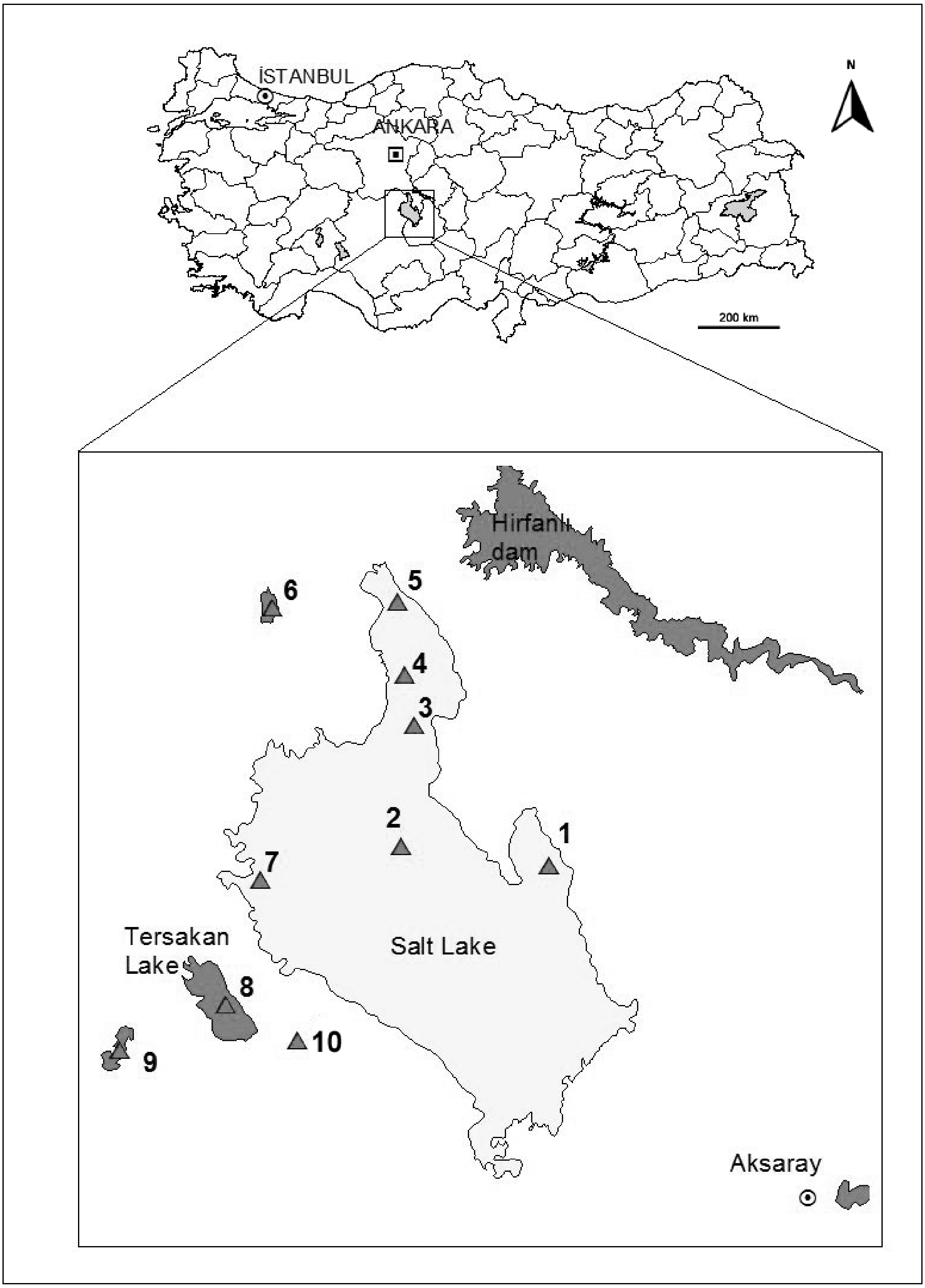


Figure 2: Sampling area and stations (1-10) in Salt Lake Basin.

RESULTS AND DISCUSSION

According to table 1, *Artemia salina* is observed only in five of these stations (1, 6, 8, 9, 10). These water bodies are: Salt Lake Aksaray, Düden Lake, Tersakan Lake, Bolluk Lake and Acı Lake. All of the water bodies where *Artemia salina* was determined are separated from the main water resources of the Salt Lake and shaped as distinctive lakes in time. *Artemia salina* was not seen in other stations.

Table 1: Measured water parameters, coordinates and individuals in 1m^3 during the study in June, July and August of 2010.

Recorded parameters	Water Level (cm)			Individuals in 1m^3			Water temperature (C°)			Electrical conductivity (mS/cm)			Dissolved Oxygen (mg/L)			pH			GPS Coordinate
Stations	Ju	Jl	Au	Ju	Jl	Au	Ju	Jl	Au	Ju	Jl	Au	Ju	Jl	Au	Ju	Jl	Au	
1. Station (Salt Lake Aksaray)	25	25	23	126	148	141	21	22	21	150	156	153	4.6	4.8	4.6	7.9	7.8	7.8	N 38°46' E 33°36'
2. Station (Salt Lake)	10	9	10	0	0	0	21	22	21	429	442	439	2.9	3.1	3.0	7.9	7.9	8.0	N 38°47' E 33°23'
3. Station (Şereflikoçhisar)	9	9	9	0	0	0	21	21	21	513	517	516	2.8	2.9	2.8	8.3	8.1	8.1	N 38°56' E 33°23'
4. Station (Salt Lake Kulu)	9	8	8	0	0	0	21	22	22	508	513	511	3.6	3.4	3.7	8.1	8.0	7.9	N 39° 0' E 33°23'
5. Station (Salt Lake Ankara)	8	8	8	0	0	0	21	22	21	494	502	501	2.8	3.0	2.9	8.0	7.8	8.0	N 39° 5' E 33°21'
6. Station (Düden Lake)	26	26	25	208	223	201	20	22	21	13	14	13	7.5	7.4	8.3	9.4	9.1	9.3	N 39° 5' E 33° 8'

Table 1 (continued): Measured water parameters, coordinates and individuals in $1m^3$ during the study in June, July and August of 2010.

Recorded parameters	Water Level (cm)			Individuals in $1m^3$			Water temperature (Cº)			Electrical conductivity (mS/cm)			Dissolved Oxygen (mg/L)			pH			GPS Coordinate
7. Station (Salt Lake Kaynak)	12	12	12	0	0	0	20	22	21	204	223	214	4.5	5.1	4.9	8.0	7.8	7.9	N 38°46' E 33°13'
8. Station (Tersakan Lake)	91	86	72	1041	1271	1132	19	21	21	148	154	150	8.9	8.6	8.7	7.9	7.8	7.9	N 38°35' E 33° 4'
9. Station (Bolluk Lake)	46	44	-	285	342	-	20	22	-	141	149	-	7.5	7.4	-	8.4	8.2	-	N 38°30' E 32°54'
10. Station (Acıgöl, Gölyazı)	28	23	22	91	147	95	20	22	21	26	33	31	4.4	4.5	4.4	8.7	8.6	8.9	N 38°31' E 33°13'

Distribution of *Artemia salina* in the Salt Lake

Artemia salina is able to live until 4500 m altitude, which is a wide range (Xin et al., 1994). The Salt Lake study environment is situated at 905 m altitude. *Artemia salina*, living in Hyper Saline waters is recorded at more than 600 coastal and inland waters (Van Stappen, 2002). One of these water areas where *Artemia salina* is found naturally as stocks, is in Van Lake, the second biggest lake of Turkey in terms of square meters; it is situated at the center of the country (between Aksaray, Ankara and Konya cities). It is reported that *Artemia salina* in Turkey is found in Ayvalık Saltpan (Koru, 2006), Çamaltı Saltpan (Koru, 2004), Salt Lake Basin (Aksaray, Ankara and Konya) (Triantaphyllidis et al., 1998) and Gökçeada (Çanakkale). In the present study, *Artemia salina* is only found in five of the stations. These stations are; Salt Lake Aksaray, Düden Lake, Tersakan Lake, Bolluk Lake and Acı Lake.

Distribution of *Artema salina* in the Salt Lake is analyzed by Başbuğ (1996) in 1995; according to the analysis of these samples collected from these stations, *Artemia salina* is found only in two stations (Devekonağı and Çalören). During the past 15 years, water levels of some water sources decreased while some lakes completely disappeared. The station named as the 5th station by Başbuğ (1996) is the same area which we named as the 1st, Salt Lake Aksaray station in our study. *Artemia salina*'s presence is determined in both studies in the area. Different from Başbuğ (1996), as in our study, new living spaces are determined where *Artemia salina* is distributed.

Abundance of *Artemia salina* in the Salt Lake

According to the data of the field study, it was determined that the number of flamingos in the 8th station, named as Tersakan Lake, where *Artemia salina* is found densely, is much higher than the other water bodies. Related with *Artemia salina* density, it is observed that the flamingo numbers in Düden and Bolluk Lakes are higher than the number of flamingos in the area which we name as the 1st station (Salt Lake Aksaray) and in Acıgöl (Gölyazı). There were no flamingos in the other five areas where *Artemia salina* is not found. This result shows us that there is a direct proportion between the *Artemia salina* density and flamingo numbers.

It is observed that the electrical conductivity in the 8th station, Tersakan Lake, is the most suitable value for *Artemia salina*; and individual number measured per cubic meter is found to be very high in this station. As known, *Artemia salina* is a very economic creature in terms of high feeding value, egg production and being used as aquarium fisheries in order to produce live bait. According to the study's result, in case of building a natural *Artemia salina* facility in future, the most suitable place is Tersakan Lake.

Relation between electrical conductivity and *Artemia salina*

At the end of measuring water parameters, we determined that in these 5 different areas in the Salt Lake, where *Artemia salina* is observed, the level of salinity content rate is much higher than the other aquatic areas. This result shows that *Artemia salina* can not accommodate in aquatic areas where salinity content rate is high. During the study, the lowest electrical conductivities were measured in July and August, at the 6th station as 13 mS/sm (Düden lake); the highest electrical conductivity was measured in June, at the 3rd station as 517 m^2/cm (Şereflikoçhisar.) The densest electrical conductivity was in July, at the 8th station (Tersakan Lake) and measured to be 154 m^2/cm. On the other hand, *Artemia salina* is not seen in the stations where electrical conductivity is 204 m^2/cm and 517 m^2/cm. In this case, 154 m^2/cm values for electrical conductivity is defined to be the most appropriate value for *Artemia salina*'s fertility. In addition to this, according to the measured values, *A. salina* can not live in 204 mS/cm or higher electrical conductivity.

Relation between water levels and *Artemia salina*

During the study, it was noticed that there are many underground water pumps in the region built for the irrigation of agricultural lands. These water pumps are used in the irrigation of agricultural lands during the summer season before waters reach the lake in the region. It is observed that water levels in the basin are very low because of the low water capacity of other water resources in the region during summer. Salinity levels increase as water heats up more during summer because of the low water levels. The increase in salinity levels create a toxic effect on *Artemia salina*. This is why, the lives of flamingos living in Salt Lake basin, whose essential nutrients is *Artemia salina*, are under threat.

Finally, the implications of these results might be valuable to balance the conservation and exploitation of natural populations of *Artemia salina* in the Salt Lake Basin in Turkey.

REFERENCES

1. Başbuğ Y., 1999 – Reproduction characteristics of Artemia salina (L., 1758) in Salt Lake, *Turkish Journal of Zoology*, 23, 2, 635-640.
2. Bengtson D., Leger P. and Sorgeloos P., 1991 – Use of Artemia as food source. In Browne, R. A., P. Sorgeloos and C.N.A. Trotman (eds), Artemia Biology. CRC Pres. Boca Raton, Florida, USA, 255-285.
3. Lengyel E., Oprean L., Tiţa O., Iancu R. and Iancu M., 2012 – Biodiversity of the microorgamisms existing in the salt lakes at Ocna Sibiului (Romania) and Chott El Jerid (Tunisia), *Transylvanian Review of Systematical and Ecological Research*, 14, The Wetlands Diversity, 25-32.
4. Koru E., 2004 – Çamaltı saltlası (İzmir, Türkiye) ekosisteminde Artemia ve önemi, *E.U. Journal of Fisheries & Aquatic Sciences*, 21, 187-189.
5. Koru E., 2006 – Ayvalık Saltlası'ndaki (Balıkesir/Türkiye) Artemia parthenogenetica'nın Yağ Asitleri Üzerine Bir Araştırma, *E.U. Journal of Fisheries & Aquatic Sciences*, 23, 185-187.
6. Triantaphyllidis G. V., Abatzopoulos T. J. and Sorgeloos P., 1998 – Review of the biogeography of the genus Artemia (Crustacea, Anostraca), *Journal of Biogeography*, 25, 213-226.
7. Xin N., Sun J., Zhang B., VanStappen G. V., Triantaphyllidis G. V. and Sorgeloos P., 1994 – International study on Artemia, New survey of Artemia resources in the People's Republic of China, *International Journal of Salt Lake Research*, 3, 105-112.
8. Van Stappen G., 2002 – Zoogeography, In Artemia: Basic and Applied Biology (eds Abatzopoulos, T. J. et al.), Kluwer, The Netherlands, 171-224.

EVALUATION OF MOSQUITO *CULEX QUINQUEFASCIATUS* MORTALITY RATE IN THE FACE OF LETHAL CONCENTRATION OF DELTAMETHRIN AND DIAZINON TOXINS

Zeinab HOSSEINNIA *, *Reza TARKHANI* **, *Fatemeh DARABITABAR* ***, *Mohmmod POORHOSENI* **** and *Aliakbar HEDAYATI* *****,

* Faculty of Fisheries and Environment, Gorgan University of Agricultural Sciences and Natural Resources, Gorgan, Iran. P. O. Box 219, IR-49316-15581, z.hosseinnia@yahoo.com
** Faculty of Natural Resources, Urmia University, Urmia, Iran., P. O. Box 795, IR-62584-69545, rezat_65@yahoo.com
*** Faculty of Marine Natural Resources- Khorramshahr University of Marine Science and Technology, Khorramshahr, Iran. P. O. Box 669, IR-66741-95326, darabitabar@gmail.com
**** Faculty of Fisheries and Environment- Gorgan University of Agricultural Sciences and Natural Resources, Gorgan, Iran. P. O. Box 219, IR-49316-15581, sm.poorhoseini@yahoo.com
***** Faculty of Fisheries and Environment- Gorgan University of Agricultural Sciences and Natural Resources, Basij Square, Daneshgah Boulevard, Gorgan, Iran. P. O. Box 219, IR-49316-15581, Hedayati@gau.ac.ir

DOI: 10.1515/trser-2017-0012

KEYWORDS: Pesticide, Aquatic Insect, LC_{50}, Pollution, Wetland

ABSTRACT

Because of huge consumption for agricultural purpose, presence of pesticides in surface waters of Golestan province (north east of Iran) is very common. These pesticides finally could be stored in aquatic ecosystems and have proven toxic effects on aquatic animals. *Culex quinquefasciatus* is one of the most common aquatic insects living mainly in the wetlands. The aim of this study was to determine the acute toxicity of deltamethrin and diazinon as potentially perilous organic pesticides and to assess mortality effects of these chemicals on the mosquito *Culex* sp. in the form of LC_{50}.

RESUME: Evaluation du taux de mortalité chez *Culex quinquefasciatus* causé par des concentrations létales de deltamethrine et de diazinone.
A cause de l'utilisation en très grandes quantités de pesticides en agriculture, ces substances sont très fréquemment présentes dans les eaux de surface de la province de Golestan (nord-est de l'Iran). Ces pesticides finissent par se déposer dans les écosystèmes aquatiques, ayant des effets toxiques prouvés sur les animaux aquatiques. Le moustique est un insecte aquatique très commun vivant notamment dans des zones humides. Le but de l'étude a été de déterminer la toxicité aigue de la deltamethrine et de la diazinone, des pesticides organiques potentiellement dangereuses, sous la forme de LC_{50}, sur le moustique *Culex quinquefasciatus***.**

REZUMAT: Evaluarea ratei de mortalitate a țânțarului *Culex quinquefasciatus* la concentrații letale ale toxinelor deltametrină și diazinonă.
Datorită utilizării în cantități uriașe în agricultură, pesticidele sunt foarte des întâlnite în apele de suprafață din provincia Golestan (nord-estul Iranului). Acestea ajung în final să se acumuleze în ecosistemele acvatice cu efecte toxice dovedite asupra faunei acvatice. *Culex* sp. este una dintre insectele acvatice cel mai des întâlnite, trăind îndeosebi în zonele umede. Scopul acestui studiu a fost determinarea toxicității acute a deltametrinei și diazinonei, două pesticide organice potențial periculoase, prin evaluarea mortalității sub forma LC_{50} induse de aceste substanțe chimice asupra speciei de țânțar *Culex quinquefasciatus*.

INTRODUCTION

There is a growing concern over aquatic contamination because of its harmful effects on biological life including human beings, chemical pesticides with continuous molecules (lengthy half-life cycles) pose a threat to various forms of aquatic life and also to the humans consuming the affected fish (Darko et al., 2008; Curtean-Bănăduc et al., 2016; Bănăduc et al., 2016; Sandu et al., 2008). *Culex pipiens* is a common mosquito that is consumed by fish. Mosquitoes are flying, biting insects that increase in water during their immature phases. It matures from egg to adult in seven days; adults generally live 10-60 days. (Langston, 1990)

Presence of pesticide in level waters was reported in North America and Europe since fifty years ago and also, many documents have demonstrated the toxic effects of these contaminants on the aquatic environment (Miller et al., 2002; Galloway and Handy, 2003; Tinoco and Halperin, 1998; Capel et al., 2001). Organophosphorus pesticides are widely used in agronomy or agriculture, and the aquatic environment near to fields is under penetration of OPs such as diazinon (Tinoco and Halperin, 1998).

Diazinon (organophosphorus pesticide) possesses moderately persistence constitution and is very used in agriculture (Bazrafshan et al., 2007; Larkin and Tjeerdema, 2000). The diazinon toxicity is due to inhibition of acetylcholinesterase activity, which causes detrimental impacts on non-aim aquatic species close to agricultural fields (Larkin and Tjeerdema, 2000).

Pyrethroids containing deltamethrin are largely used as pediculicides and are among the most potent insecticides (Viran et al., 2003). Pyrethroids are extremely toxic to shrimps and fish and some aquatic arthropods (Viran et al., 2003; Srivastav et al., 1997).

The acute toxicity information provides useful data to determine the action mode and comparison of the amount response among different chemicals. 96-h LC_{50} experiments are conducted to determine the survival potential of organisms to particular poisonous chemicals. Chemicals with lower LC_{50} amount are more toxic because lower concentrations result in 50% of fatality or mortality in organisms.

MATERIAL AND METHODS

The studied species was *Culex quinquefasciatus*. Lethal experiments were conducted using 100 individuals. Test chambers were Petri dish. All Culexes were adapted for 2 days in these aquaria before assays. Water temperature was regulated at 27°C. Dead Culex were immediately removed to avoid possible water pollution (Gooley et al., 2000).

Substantial concentrations of active ingredient tested were 0, 1, 4, 10, 20, 40, 100 and 200 ppm of commercial dose (60%) for diazinon and 0, 1, 4, 10, 20, 40, 100 and 200 ppm of commercial dose (2.5%) for deltamethrin. 14 groups (seven for diazinon and seven for deltamethrin) of 100 mosquitos were exposed for 96 hours in the Petri dishes.

During the acute toxicity experiment, the water temperature in Petri dish was 27°C. There was not food administered during the assay and test media was not renewed. Mortality rates were recorded at 24, 48, 72 and 96 h. Acute toxicity tests carried out according to Hotos and Vlahos (Hotos and Vlahos, 1998). Nominal concentration of diazinon and deltamethrin in 50% mortality of *Culex* mosquito was attained by probit analysis in Finney's method within 24 h, 48 h, 72 h, and 96 h (Finney, 1971) in the maximum-likelihood procedure (SPSS 2002, SPSS Inc., Chicago, Illinois, USA). Lethal concentration value was obtained by graphical interpolation by taking logarithms of deltamethrin and diazinon concentrations versus probit value of percentage mortality and also by fitting a regression equation arithmetically.

A 95% confidence limits for LC50 were computed by using the formula:

$$(LC_{50}\ 95\%\ CL) = LC50 \pm 1/96\ [SE\ (LC_{50})]$$

The SE of LC_{50} was determined with formula: $SE(LC50) = 1/b\sqrt{pnw}$

Where: b = the gradient of the chemical-probit analyser response (regression) line; w = the average weight of the observations, p = the number of chemical used, n= the number of animals in each group (Hotos and Vlahos, 1998). After the acute toxicity test, the NOEC (No Observed Effect Concentration) and LOEC (Low Observed Effect Concentration) were calculated for each measured endpoint.

RESULTS AND DISCUSSION

There was no mortality during the acclimation period before exposure, also there was no mortality in the control group during acute toxicity tests. The mortality of mosquito *Culex quinquefasciatus* for both of deltametrin and diazinon are in table 1 and 2 during the exposure times at (24-96 h) respectively. The mortality of mosquito *Culex quinquefasciatus* increased significantly with increasing concentrations (from 1 ppm to higher concentrations) for both of diazinon and deltamethrin.

However for both of deltamethrin and diazinon there were 100% mortality at 200 and higher concentrations within the 96 h after exposure, and no observed 100% mortality within the 24 h (Tabs. 1 and 2).

Table 1: Cumulative mortality of Mosquito *Culex quinquefasciatus* (n=100 each concentration) exposed to acute diazinon.

Concentration (ppm)	No. of mortality			
	24 h	48 h	72 h	96 h
0	0	0	0	0
1	0	0	0	6
4	0	2	8	30
10	0	3	15	45
20	0	3	17	48
40	3	10	29	59
100	15	40	73	89
200	43	62	100	100

Table 2: Cumulative mortality of Mosquito *Culex quinquefasciatus* (n = 100, each concentration) exposed to acute deltamethrin.

Concentration (ppm)	No. of mortality			
	24h	48h	72h	96h
0	0	0	0	0
1	0	0	2	4
4	0	1	5	10
10	15	31	53	61
20	37	49	64	73
40	49	63	69	82
100	61	83	88	97
200	80	89	100	100

Median lethal concentrations of (10-90%) experiment are in tables 3 and 4. Because mortality (or survival) information were prepared for each exposure concentration in a toxicity test at different exposure durations (24, 48, 72, or 96 hours), data can be designed in other ways; the direct line of best fit is then drawn through the spots. These are time-mortality lines. The LT_{50} (median lethal survival time) can be computed for each concentration.

Table 3: LC50 (1-99) of diazinon depending on time (24, 48, 72 and 96h) for Mosquito *Culex quinquefasciatus*.

POINT	CONCENTRATION (PPM) (95 % OF CONFIDENCE LIMITS)			
	24h	48h	72h	96h
LC_1	18.8 ± 0.01	-	-	-
LC_{10}	103.3 ± 0.01	49.9 ± 0.01	9.8 ± 0.02	-
LC_{20}	138.8 ± 0.01	86.6 ± 0.01	29.9 ± 0.02	-
LC_{30}	164.5 ± 0.01	113.0 ± 0.01	44.4 ± 0.02	6.5 ± 0.02
LC_{40}	186.4 ± 0.01	135.5 ± 0.01	56.7 ± 0.02	19.6 ± 0.02
LC_{50}	**206.9 ± 0.01**	**156.6 ± 0.01**	**68.3 ± 0.02**	**31.9 ± 0.02**
LC_{60}	227.4 ± 0.01	177.7 ± 0.01	79.8 ± 0.02	44.2 ± 0.02
LC_{70}	249.3 ± 0.01	200.2 ± 0.01	92.2 ± 0.02	57.3 ± 0.02
LC_{80}	274.9 ± 0.01	226.6 ± 0.01	106.6 ± 0.02	72.7 ± 0.02
LC_{90}	310.5 ± 0.01	263.3 ± 0.01	126.7 ± 0.02	94.0 ± 0.02
LC_{99}	395.0 ± 0.01	350.2± 0.01	174.3 ± 0.02	114.5±0.02

Table 4: LC50 (1-99) of deltamethrin depending on time (24, 48, 72 and 96 h) for Mosquito *Culex quinquefasciatus*.

POINT	CONCENTRATION (PPM) (95 % OF CONFIDENCE LIMITS)			
	24h	48h	72h	96h
LC_1	-	-	-	-
LC_{10}	-	-	-	-
LC_{20}	17.7 ± 0.01	-	-	2.7 ± 0.04
LC_{30}	46.2 ± 0.01	19.3 ± 0.01	8.3 ± 0.02	5.2± 0.04
LC_{40}	70.6 ± 0.01	39.3 ± 0.01	19.7 ±0.02	12.1±0.04
LC_{50}	**93.4 ± 0.01**	**58.1 ± 0.01**	**30.3 ± 0.02**	**18.5 ±0.04**
LC_{60}	116.2 ±0.01	76.8 ± 0.01	40.9 ±0.02	24.9± 0.04
LC_{70}	140.6 ±0.01	96.9 ± 0.01	52.2 ±0.02	31.8 ±0.04
LC_{80}	169.2 ±0.01	120.3 ±0.01	65.5 ±0.02	39.8 ±0.04
LC_{90}	208.8 ±0.01	152.0 ±0.01	83.9 ±0.02	50.9 ±0.04
LC_{99}	302.8 ±0.01	230.1 ±0.01	127.7±0.02	77.4 ±0.04

Toxicity Testing Statistical Endpoints are presented in figures 1 and 2.

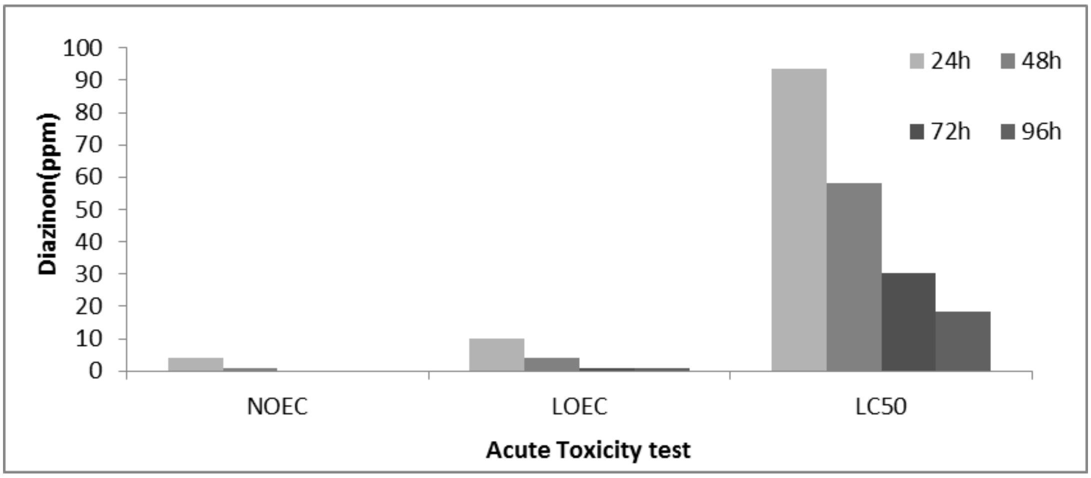


Figure 1: Acute toxicity experimenting statistical endpoints in *Mosquito Culex quinquefasciatus* exposed to Diazinon at different times (24-96 h).

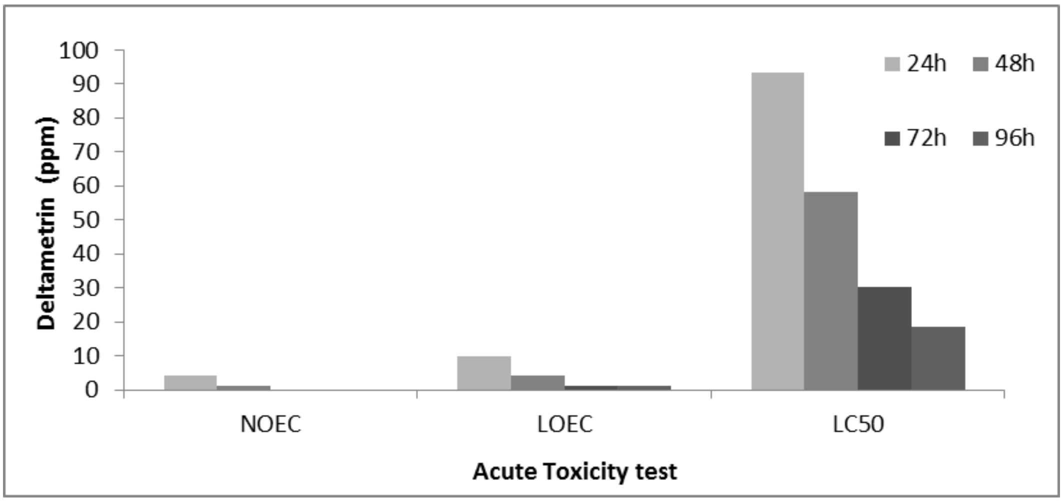


Figure 2: Acute toxicity experimenting statistical endpoints in exposed to deltamethrin at different times (24-96 h).

Incidence of pesticides in high concentrations in agricultural wastewater and their toxicity to aquatic organisms such as fish species have been reported by many researchers (Galloway and Handy, 2003; Capel et al., 2001; Larkin and Tjeerdema, 2000). The effects of the insecticide diazinon and thiacloprid at temperatures 28, 30 and 33.5°C on the growth of zebra fish eggs *Danio rerio*, were examined. Results have shown that the toxic effect of diazinon increases further with increasing temperature, (Heinz, 2008). Chakabarty and Banergee (1988) calculated and reported the LC_{50} concentration for diazinon granules equal to 5 mg per liter (5%) (Chakabarty and Banergee, 1988). Wege et al reported a 99% mortality for adult male German cockroaches exposed to 30 mg per square meter of active ingredient wettable powder formulation Lambda cyhalothrin on metal surfaces after 30 seconds. The researchers formulation of microcapsules in the same concentration resulted in a mortality of 95%. The metal surface is a nonabsorbent surface and caused many casualties after 30 seconds of contact with the insect while it develops (Wege, 1999).

Contamination of water organisms and environment with pesticides via rainfall runoff is very conceivable (Willis and McDowell, 1982). In addition it found that both diazinon and deltamethrin were lethal substrates to mosquito. The mortality of mosquito increased significantly with increasing concentrations (from 1 ppm to higher concentrations) for both of diazinon and deltamethrin. However for both of deltametrin and diazinon there was 100% mortality at 200 and higher concentrations within the 96h after exposure, and no observed 100% mortality within the 24 h. The 96h LC_{50} was calculated to be 31.9 ± 0.02 ppm for diazinon and 18.5 ± 0.04ppm for deltamethrin and here it reported that diazinon was lowly toxic to mosquito *Culex quinquefasciatus*.

LC50 values of aqueous and organic solvent extracts of plants/plant parts on mosquito species reported from tenths to several tens of mg l^{-1} (Raghavendra, 2009). Value of diazinon 96h LC_{50} was 0.8 mg/l for zebra fish (*Brachydanio rerio*) and it was 8 mg/l for guppy (*Poecilia reticulata*) (Adedeji et al., 2008; Keizer et al., 1991). Getaway reaction of unmated and mated nulliparous *Aedes aegypti* mosquitoes were estimated using three different concentrations of deltamethrin in the presence or absence of a live organism host (Boonyuan et al., 2011). Although deltamethrin is thought to be lower toxic in field conditions due to its adsorption to sediment, these data are useful to potential ecosystem risk evaluation (Viran, 2003).

CONCLUSIONS

Our results showed that LC_{50} 96 h of mosquito *Culex quinquefasciatus* was calculated to be 31.9 ppm for diazinon and 18.5 ppm for deltamethrin. Maximum allowable concentration was 3.19 mg/l for diazinon and 1.85 mg/l deltamethrin respectively. The results of the present study suggest that both chemicals diazinon and deltamethrin varied in their acute toxicity on the mosquito. The toxicity of deltamethrin and diazinon on the mosquito increased with increasing the concentration and exposure time.

ACKNOWLEDGEMENTS

This work was supported by the Gorgan University of Agricultural Sciences and Natural Resources.

REFERENCES

1. Adedeji O. B., Adedeji A., Adeyemo O. K. and Agbede S. A., 2008 – Acute toxicity of diazinon to the African catfish (Clarias gariepinus), *African Journal of Biotechnology,* 7, 5, 651-654.
2. Bazrafshan E. S., Naseri A .H., Mahvi M. and Shayedhi , 2007 – Performance evaluation of electrocoagulation process for diazinon removal from aquaeous environments by using iron electrons, *Iranian Journal of Environmental Health Science and Engineering,* 4, 127-132.
3. Bănăduc D., Moza M. I., Burcea A. and Curtean-Bănăduc A., 2016 – Persistent organic pollutants in continental aquatic ecosystems, in Curtean-Bănăduc A. (ed.), The Impact of persistent organic pollutants on freshwater ecosystems and human health, 95-108, Ed. "Lucian Blaga" University of Sibiu, ISBN 978-606-12-1412-9, 154.
4. Boonyuan W., Kongmee M., Bangs M., Prabaripai A. and Chareonviriyaphap T., 2011 − Host feeding responses of *Aedes aegypti* (L.) exposed to deltamethrin, *Journal of Vector Ecology,* 36, 2, 361-372.
5. Capel P. D., Larson S. J. and Winterstein T. A., 2001 − The behavior of thirty-nine pesticides in surface waters as a function of scale, *Hydrologica Process,* 15, 1251-1269.
6. Chakabarty P. and Banergee V., 1988 − Effect of sublethal toxicity of three organophosforus pesticides on the peripheral hemogrm of the fish Channa punctatus, *Enviroment and Ecology,* 151-158.
7. Curtean-Bănăduc A., Lyche J. L., Berg V., Burcea A. and Bănăduc D. 2016 – Assessment and monitoring of persistent organic pollutants in lotic ecoszstems – methodological guide, publisher "Lucian Blaga" University of Sibiu,ISBN 978-606-12-1414-3, 117.
8. Darko G., Akoto O. and Oppong C., 2000 – Persistent organochlorine pesticide residues in fish, sediments and water from Lake Bosomtwi, Ghana, *Chemosphere*, 72, 21-24.
9. Finney D. J., 1971 − Probit Analysis, University Press, Cambridge, 333.
10. Galloway T. and Handy R., 2003 − Immunotoxicity of organophosphorous pesticides, *Ecotoxicology,* 12, 345-63.
11. Gooley G. J., Gavine F. M., Dalton W., De Silva S. S., Bretherton M. and Samblebe M., 2000 − Feasibility of aquaculture in dairy manufacturing wastewater to enhance environmental performance and offset costs, Final Report DRDC Project No. MAF001, Marine and Freshwater Resources Institute, Snobs Creek, 84.
12. Heinz R. K., 2008 – Pesticides thiacloprid and diazinon on the embryonic development of zebrafish (Danio rerio), Academic Press, 311.
13. Hotos G. N. and Vlahos N., 1998 − Salinity tolerance of *Mugil cephalus* and *Chelon labrosus,* Pisces: Mugilidae/fry in experimental conditions, *Aquaculture*, 167, 329-338.
14. Keizer J. D., Gostino G. and Vittozzi L., 1991 − The importance of biotransformation in the toxicity of xenobiotics to fish.1, Toxicity and bioaccumulation of diazinon in guppy (Poecilia reticulata) and zebra fish (Brachydanio rerio), *Aquatic Toxicology,* 21, 239-254.
15. Langston W. J., 1990 − Toxic effects of metals and the incidence of metal pollution in marine ecosystems. In: Firness, R.W; Rainbow, P.S. (Eds.), Heavy Metals in the Marine Environment. CRC Press, Boca Raton, FL, 101-122.
16. Larkin D. J. and Tjeerdema R. S., 2000 − Fate and effects of diazinon, *Reviews in Environmental Contamination and Toxicology,* 166, 49-82.
17. Miller G. G., Sweet L. I., Adams J. V., Omann G. M., Passino-Reader D. R. and Meier P. G., 2002 − In vitro toxicity and interactions of environmental contaminants (Arochlor 1254 and mercury) and immunomodulatory agents (lipopolysaccharide and cortisol) on thymocytes from lake trout (Salvelinus namaycush), *Fish and Shellfish Immunology,* 13, 11-26.
18. Raghavendra K., Singh S. P. and Sarala K., 2009 − Subbarao and Dash, A. P. Laboratory studies on Mosquito larvicidal efficacy of aqueous and hexane extracts of dried fruit of Solanum nigrum Linn, *Indian Journal of Medical Research*, 130, 74-77.
19. Sandu C., Bloesch J. and Coman A., 2008 – Water pollution in the Mureş Catchment and its impact on the aquatic communities, *Transylvanian Review of Systematical and Ecologicak*

Research, 6, The Wetlands Diversity, 97-108.

20. Srivastav A. K., Srivastava S. K. and Srivastav S. K., 1997 − Impact of deltamethrin on serum calcium and inorganic phosphate of freshwater catfish, Heteropneustes fossilis, *Bulletin of Environmental Contamination and Toxicology*, 59, 841-846.
21. Tinoco-Ojanguren R. and Halperin D. C., 1998 − Poverty production, and health: inhibition of erythrocyte cholinesterase via occupational exposure to organophosphate insecticides in Chiapas, Mexico, *Archive of Environmental Health*, 53, 29-35.
22. Viran R., Erkoc F. U., Polat H. and Kocak O., 2003 − Investigation of acute toxicity of deltamethrin on guppies (Poecilia reticulata), *Ecotoxicology and Environmental Safety*, 55 , 82-85.
23. Wege P. J., Hoppe M. A., Bywater A. F., Weeks S. D. and Gallo T. S., 1999 − A microencapsulated formulation of lambda-cyhalothrin, Proceedings of the 3rd International Conference on Urban Pests, 91-94.
24. Willis G. H. and McDowell L. L., 1982 − Review: Pesticides in agricultural runoff and their effects on downstream water quality, *Environmental Toxicology and Chemistry*, 1, 267-279.

IMPROVEMENT OF LATERAL CONNECTIVITY IN A SECTOR OF RIVER HÂRTIBACIU (OLT/DANUBE BASIN)

Răzvan VOICU *, *Doru BĂNĂDUC* **, *Eric KAY* ***
Erika SCHNEIDER-BINDER **** and *Angela CURTEAN-BĂNĂDUC* **

* National Institute of Hydrology and Water Management, Bucureşti-Ploieşti Street 97, Bucureşti, Romania, rzvnvoicu@yahoo.com, getiiliberi@gmail.com
** "Lucian Blaga" University of Sibiu, Applied Ecology Research Center, Dr. Ion Raţiu Street 5-7, Sibiu, Sibiu County, Romania, RO-550012, angela.banaduc@ulbsibiu.ro, ad.banaduc@yahoo.com
*** Kay and Associates, 8712 Island Hwy, Black Creek, B.C., Canada, V9J 1K5, erickay@kayassociates.com
**** Karlsruhe Institute for Technology – University of Land Baden-Württemberg and Research Center of the Helmholtz Society, Institute for Geography and Geoecology, Department WWF-Institute for Floodplain Ecology, Josefstr. 1, Rastatt/ Germany, D-76437, erika.schneider@kit.partner.edu; erika.schb@t-online.de

DOI: 10.1515/trser-2017-0013

KEYWORDS: Alţâna locality, Romania, Hârtibaciu lotic system, conservation, *Rhodeus sericeus*, *Anodonta cygnea*.

ABSTRACT

The Hârtibaciu River in the Alţâna area has a disturbed lateral connectivity owing to its lateral embankments, as a result of which fish communities in this river sector are negatively influenced. The method of obtaining the water supply for a new proposed wetland is by gravitation, and any excessive water supply to the wetland will be controled and directed to the Hârtibaciu River in a natural manner. A state-of-the-art man-made wetland should increase the quality of the habitat for local fish communities, especially for the *Rhodeus sericeus amarus* population, which is a species of conservation interest.

ZUSAMMENFASSUNG: Die Verbesserung der lateralen Konnektivität in einem Abschnitt des Harbach-Flusses/Einzugsgebiet Alt/Donau.

Der Harbach hat bedingt durch die Seitendämme in Alzen/Alţâna eine gestörte laterale Konnektivität, wodurch die Fischgemeinschaft in diesem Abschnitt negativ beeinflusst wird. Die Methode für ein neu anzulegendes Feuchtgebiet die Wasserversorgung zu sichern, wird über Gravitation gelöst, wobei der Überschuss an eingeleitetem Wasser kontrolliert und auf natürlichem Weg wieder dem Harbach zugeführt wird. Ein auf dem neusten Stand des Wissens angelegtes Feuchtgebiet soll die Lebensraumqualität für die lokale Fischgemeinschaft verbessern, insbesondere für die Population von *Rhodeus sericeus amarus*, eine Art von besonderem Naturschutz fachlichem Interesse.

REZUMAT: Îmbunătăţirea conectivităţii laterale într-un sector al râului Hârtibaciu (Bazinul Olt/Dunăre).

Râul Hârtibaciu în zona Alţâna are o conectivitate laterală deteriorată datorită îndiguirilor fapt care influenţează negativ comunitatea de peşti din acest sector de râu. Metoda de a da sursă de apă unei zone umede nou propuse este realizată prin gravitaţie, iar surplusul de debit de apă din zona umedă va fi controlat şi dirijat spre râul Hârtibaciu într-un mod natural. O zonă umedă nouă, conectată cu râul trebuie să crească calitatea habitatului pentru comunităţile de peşti locale, în special pentru poluaţia de *Rhodeus sericeus amarus* - specie de interes conservativ.

INTRODUCTION

The rivers and streams connectivity, and the connectivity between them and their floodplains increases the preservation of areas for the needed specific habitat, tempers a variety of types of elements which affect the dynamics of ecosystems, improves the access to the subteranean water flows, improves the quality of water and soil, expands the depository potential for beneficial sediments, increases the flux of nutrients, depletes flood energy dispersion, rejuvenates floodplain and riparian biocoenoses and provides the ecosystem sustainability. (Gumpinger and Scheder, 2008; Ickes et al., 2005; Wilgen et al., 1998; Bănăduc et al., 2016; Hapciuc et al., 2016; Jeeva et al., 2011; Hoancă et al., 2014)

In EU, the Water Framework Directive decided modern guidelines for water, with the purpose of avoiding the deterioration of aquatic and semi-aquatic ecosystems. Only if the natural water course is managed within an integrated continuous manner, an optimum hydro-morphological and ecological dynamic can be obtained (Biswas, 2008).

Figure 1a: Human impact on the Hârtibaciu River studied sector on banks and riverbed.

In the researched area, the connectivity between the river and its floodplain was diagnosed as a needed achievable result, to support proper environment circumstances that would meet the objectives of re-establishing lateral connectivity and preventing the related risks, to all-encompassing habitats and biocoenoses, of which the fish populations belong.

For this area which is under anthropic impact it is also necessary to restore the river, the adjacent floodplain and the connectivity between them as much as possible, with the proposed solutions being based on scientific adapted concepts (Voicu and Dominguez, 2016). Disrupted fish migration to wetlands and over cross hydraulic constructions are for design engineers a scientific challenge to be solved (Kay and Voicu, 2013). Limiting rivers only to minor river bed due to various hydraulic works, agricultural works, and industrial areas and construction of houses near rivers affects the functioning of these lotic ecosystems (Voicu and Voicu, 2015).

The projected wetland is necessary for fish communities in a zone where the lotic system was channelled in the past, and incorrect activities (river bed and river banks cleaning with heavy machines, destruction of the riverine vegetation, etc.) was performed probably due to the lack of proper management expertise (Fig. 1a, b). This zone is dissimilar from the upstream semi-natural lotic sector where samplings were done (Fig. 2).

Figure 1b: Human impact on the Hârtibaciu River studied sector on banks and riverbed.

Figure 2: Semi-natural sector of Hârtibaciu River in the studied sector.

MATERIAL AND METHODS

To evaluate the necessity for the suggested project the fish association structure was researched in the Alţâna locality (Sibiu County, Romania) proximity of the river Hârtibaciu.

In 2015-2016, a mountain fishing net was used for fishing, in time and effort unit, the captured fish were identified and released back into their habitat at once.

RESULTS AND DISCUSSION

Local fish association risks and necessity for restoration of habitat

The fish of river Hârtibaciu contain a rather high number of species of fish: chub, bleak, schneider, European bitterling, gudgeon, Kessler's gudgeon, Mediterranean barbel, Stone loach, Weatherfish, Spined loach, Romanian loach, and Golden spined loach.

At the river Hârtibaciu near the Alțâna locality, a total of five fish species were sampled, namely: chub, schneider, Common nase, European bitterling, and gudgeon. In this research period, Common nase was found for the first time in the Hârtibaciu River.

The formation of the suggested wetland would offer a significant buffer zone for the fish of the area, particularly in the cold and dry periods.

In the researched lotic sector of the river Hârtibaciu, five fish species were sampled: Schneider with a relative abundance of 40%, European bitterling 28%, gudgeon 20%, chub 8%, and Common nase 4%.

The suggested new wetland should be good for the swan mussel *Anodonta cygnea* species which is living in the researched area sector. This species offer a symbiotic support for the European bitterling species breeding (Bănărescu and Bănăduc, 2007) and also for their local aquatic self-cleaning mechanism of filter feeding on planktonic algae.

It is relevant to highlight the fact that the European bitterling is a species protected under the Habitats Directive (92/43/EEC). A new wetland area can grow its abundance in the studied area as it offers a characteristic habitat with stagnant and/or semi-stagnant water with smooth type of sediment bottom (Bănărescu and Bănăduc, 2007).

Ecological reconstruction and new wetland area development formation

In present day the Alțâna Village floodplain tight near the Hârtibaciu River is used for farming and the land is privately owned.

It is therefore suggested the development of some wetland (Fig. 1) downstream of the bridge (Fig. 2a and b) whose water supply can be achieved in one of two ways by redirecting water from the Hârtibaciu River gravitationally: a) through a rectangular canal set upstream from the bridge (~ 20 m) or b) through an existing lateral canal.

The creation of the suggested new wetland area can provide a buffer area for the local fish species, and an accessible passage for their movement. The wetlands related birds, amphibians, molluscs, and vegetation – in particular riparian gallery-like forests and floodplain meadows – can also profit by this wetland, biodiversity will continue to mature, becoming more diverse, balanced and self-sustainable as pioneer species give way to evolving flora. This project can serve as an example for further projects concerning the restoration of connectivity between the river and its floodplain on a larger scale in which it can also be included that the concept of more hydrological and morphological dynamic is characteristic for natural and semi natural rivers.

Figura 1: Location of proposed wetland (www.google.com)

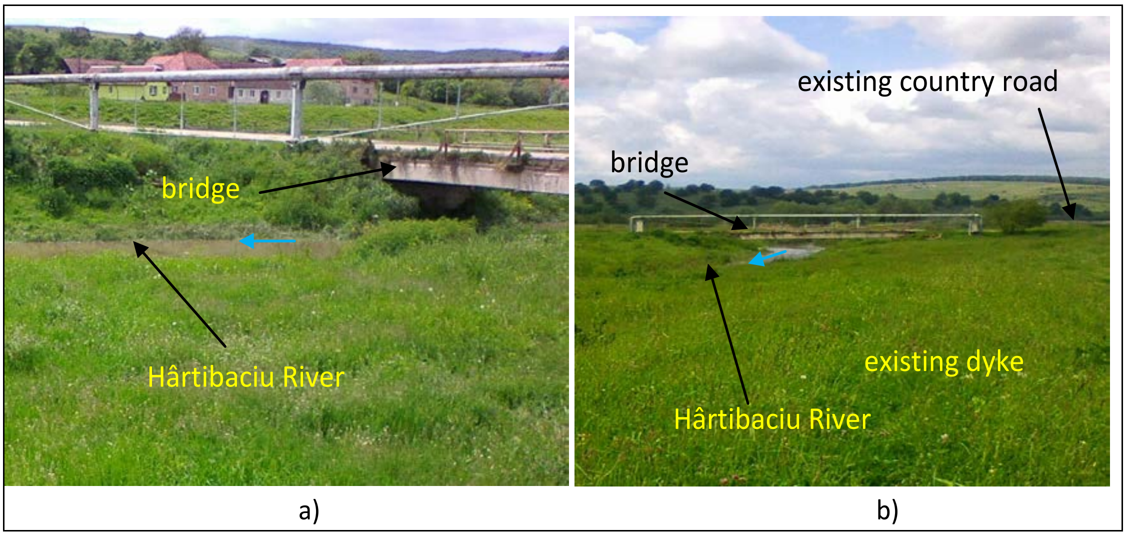


Figure 2: The Hârtibaciu River in Alțâna village, downstream of the bridge.

Steps in creating the new anthropogenic wetland.

In a first stage it is suggested to set up the study area as follows.

The entire selected future wetland perimeter is deepened by about 2.5 m in relation to the current height, while the current banks are linearized and are inclined (Fig. 3a, b). The water level in the new created wetland must be ~ 1 m, with a number of deeper pools to create a natural sanctuary habitat.

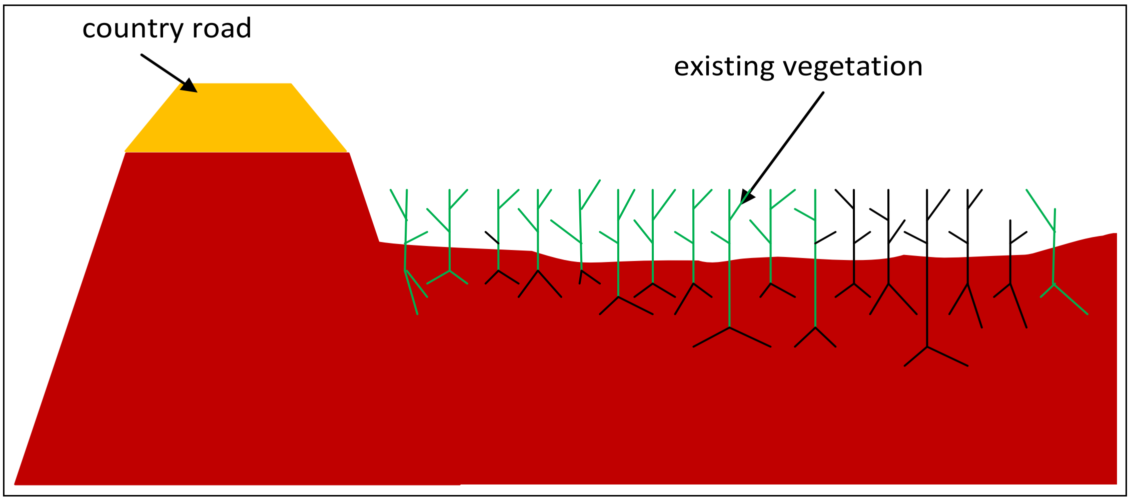


Figure 3a: Current structure of the wetland – indicative scheme.

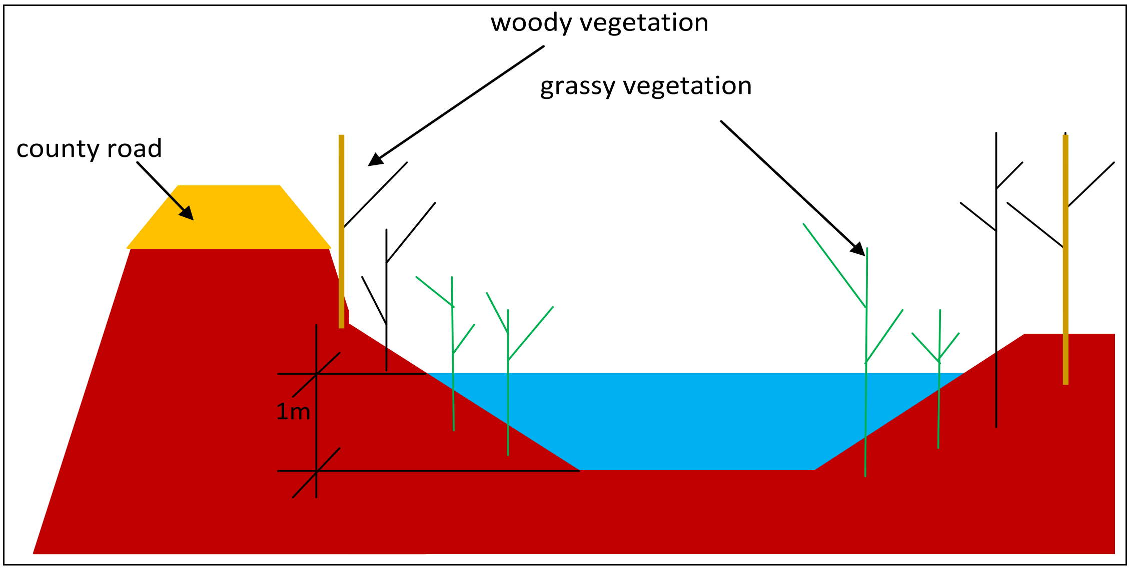


Figure 3b: Restored structure of the wetland – indicative scheme.

On the banks of the basin/wetland, planting of fast-growing woody species (black alder *Alnus glutinosa* and local characteristic willow species such are *Salix alba* and *Salix fragilis*, etc.) is recommended. The roots of these species contribute very quickly for the stability of the bank with minimal technical works. In the same time between the extended roots will develop niches, i.e. hiding places for fish species and as well for site typical macrozoobenthos. On the other hand, it is not recommended to plant either high species, as they may stagger under the wind force thus favouring the landfall, or species of herbaceous plants with tap root as it facilitates the slope stability. The planting of borders of tall herbaceous species and other herbaceous species is not needed because, they will colonise according to our experience in the course of time. But taking into account, that in the surrounding area, there exists riparian fast growing neophytes, an initial planting of site typical species would be necessary, to stop the colonisation by invasive species such are *Helianthus decapetalus*, *Rudbeckia laciniata* and *Solidago canadensis*.

However, it is recommended to plant species with fascicular roots (ex.: sedge, fescue, etc.) as they prefer moisture soil that can eliminate large amounts of water. On the first created half of bank/inclined plane, it is recommended to plant water dependent species (reed, sedge, fescue, etc.) and on the other half of the inclined plane some woody vegetation (silver poplars, black alders, willow species, etc.) (Fig. 3b). This abundant vegetation growing on the banks of river/wetland will provide habitat for various species of aquatic birds and other aquatic organisms, including fish. This vegetation also has a positive indirect and direct trophic influence for fish (Curtean-Bănăduc et al., 2014). It would be useful to include in the study also the stretch of the Hartibaciu River around the study area, including a stretch upstream and downstream to be also restored by planting typical riparian vegetation (willow species) as it exists on the Hârtibaciu River more upstream but in representative galleries as exhibited in some downstream sectors (Fig. 4)

Figure 4: Representative galleries like riverine trees vegetation.

An important aspect when creating a wetland is represented by the *continuous water supply system* of the wetland. Therefore, within this solution two methods/systems for supplying water to the wetland are proposed, as described below.

First water supply system of the wetland

It is recommended for water to be gathered from the Hârtibaciu River in a canal situated upstream of the bridge (~ 20m), using some metal sheet piling arranged inside the river. If it would be possible on this canal also it would be good to plant some willows, to increase the ecological value of these technical works and to create on the canal border some small niches for macrozoobenthos and fish species. The upper end of this rectangular canal will be placed near the sheet pile parallel to the left bank of the river (Fig. 5) whilst the lower end of this rectangular canal will be placed near the wetland (Fig. 6). At the upstream end of the canal there will be set a metal grille (Fig. 6) blocking the floating elements and a metal sluice with manual aperture that help maintain this canal and repair the entire water supply system of the wetland.

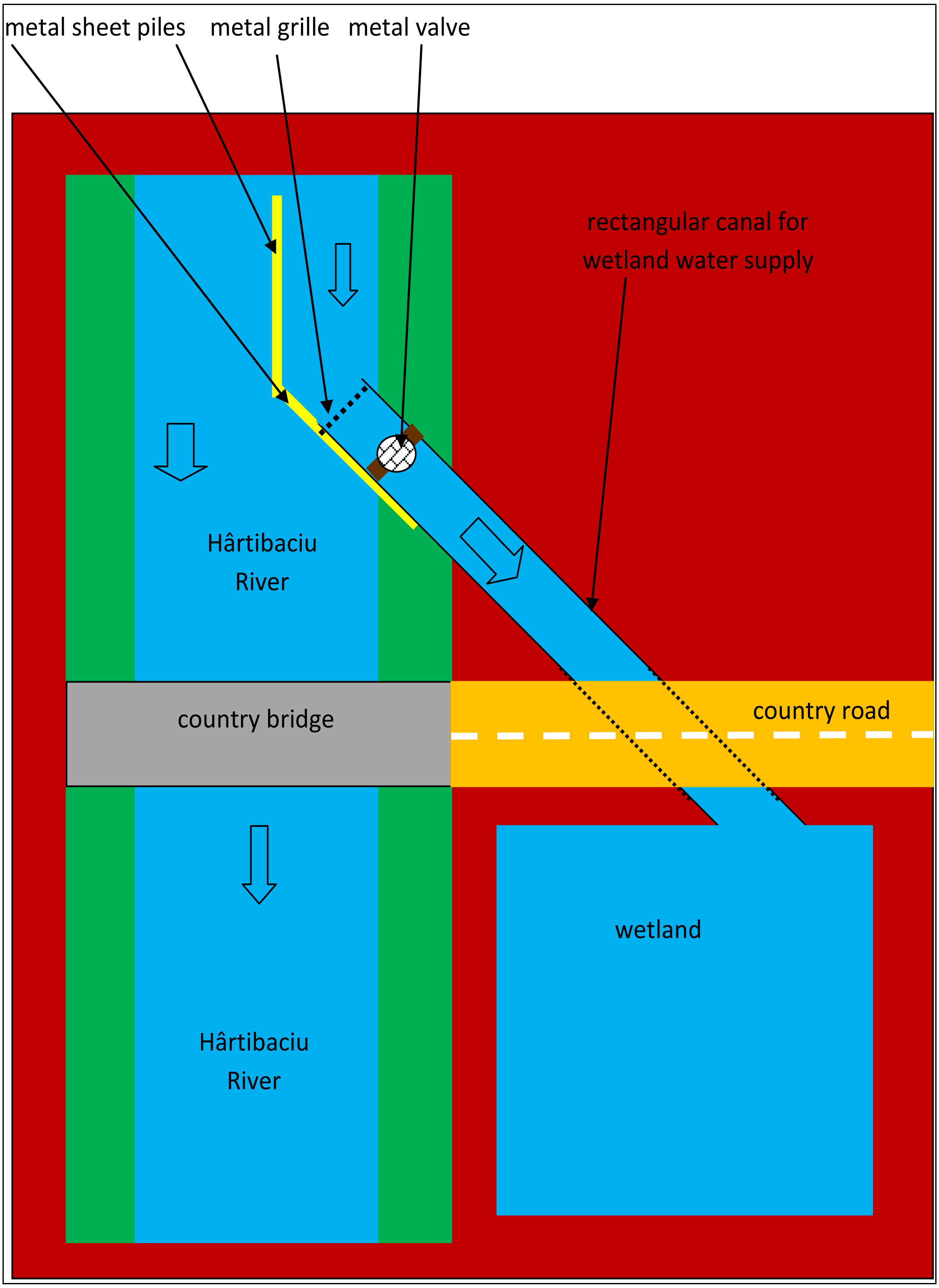


Figure 6: Positioning water supply canal of the wetland
– indicative scheme.

Wetland water supply is realized gravitationally by the means of the rectangular canal. To avoid the erosion of this canal it is recommended to strengthen it by using some wooden bars (treated against moisture) and arranged all over its length (Fig. 7).

The difference between the rectangular channel supplying the wetland and the water surface is one meter. Water discharge into the wetland will be achieved also by a rectangular controlled channel having the same dimensions as the channel for wetland water supply (Fig. 8).

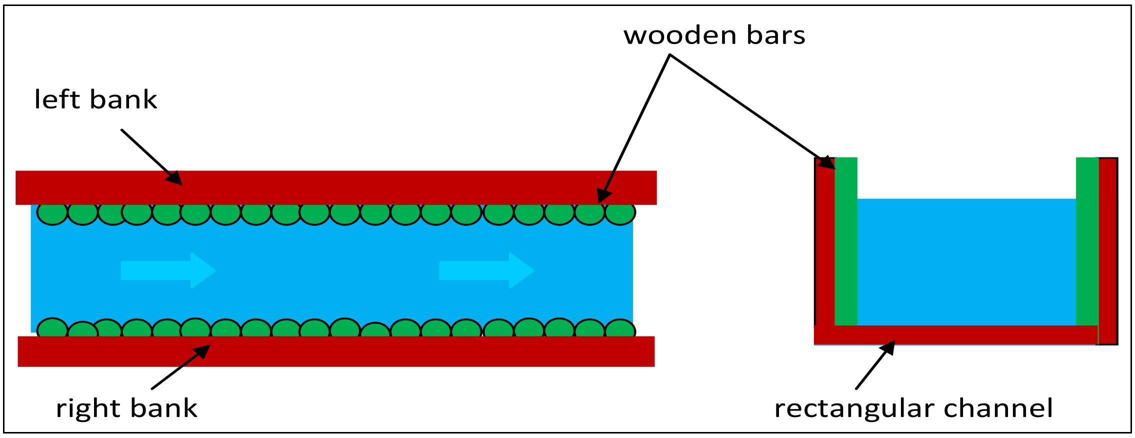


Figure 7: Strengthening the rectangular channel by using some wooden bars – indicative scheme.

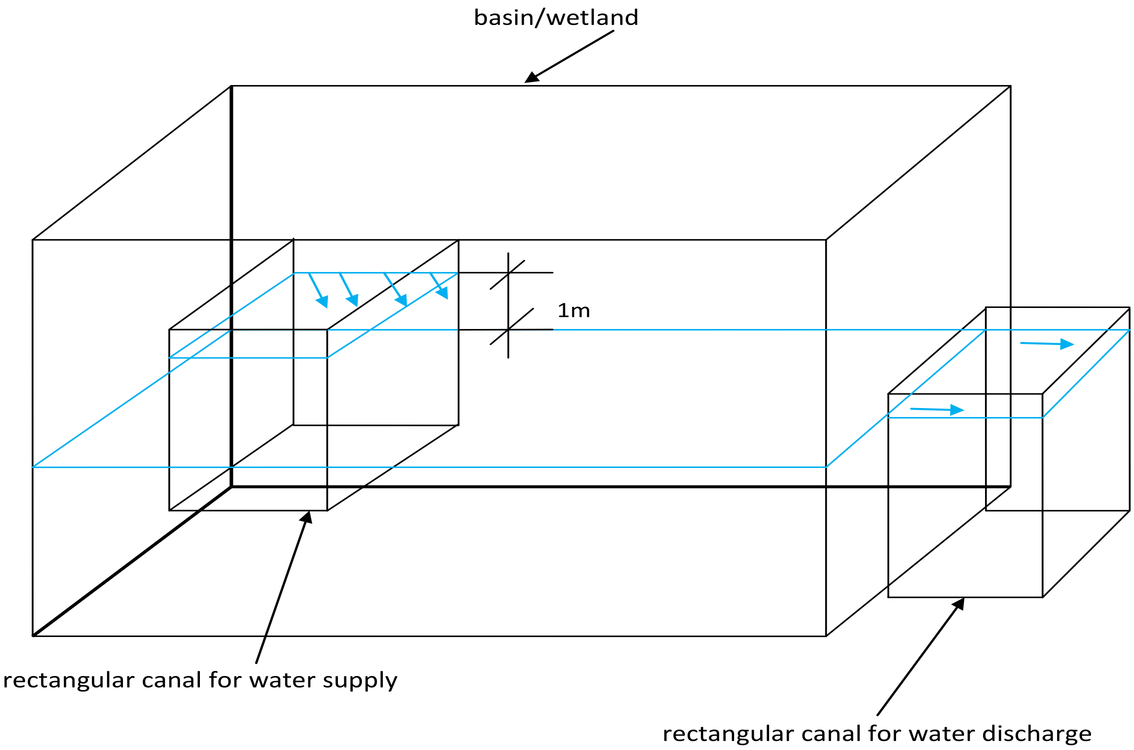


Figure 8: Positioning the canal for water discharge – indicative scheme.

Because of the water discharge canal, the water level in the created wetland is 1 m constantly. Also, the water discharge canal will be arranged like the one for water supply as regarding the banks strengthening. Thus the water discharge into the wetland is achieved gravitationally and directly into the Hârtibaciu River (Fig. 9). No more than 300m away from the wetland, the Hârtibaciu River loses 40% of its maximum rate which actually does not affect the local fish fauna.

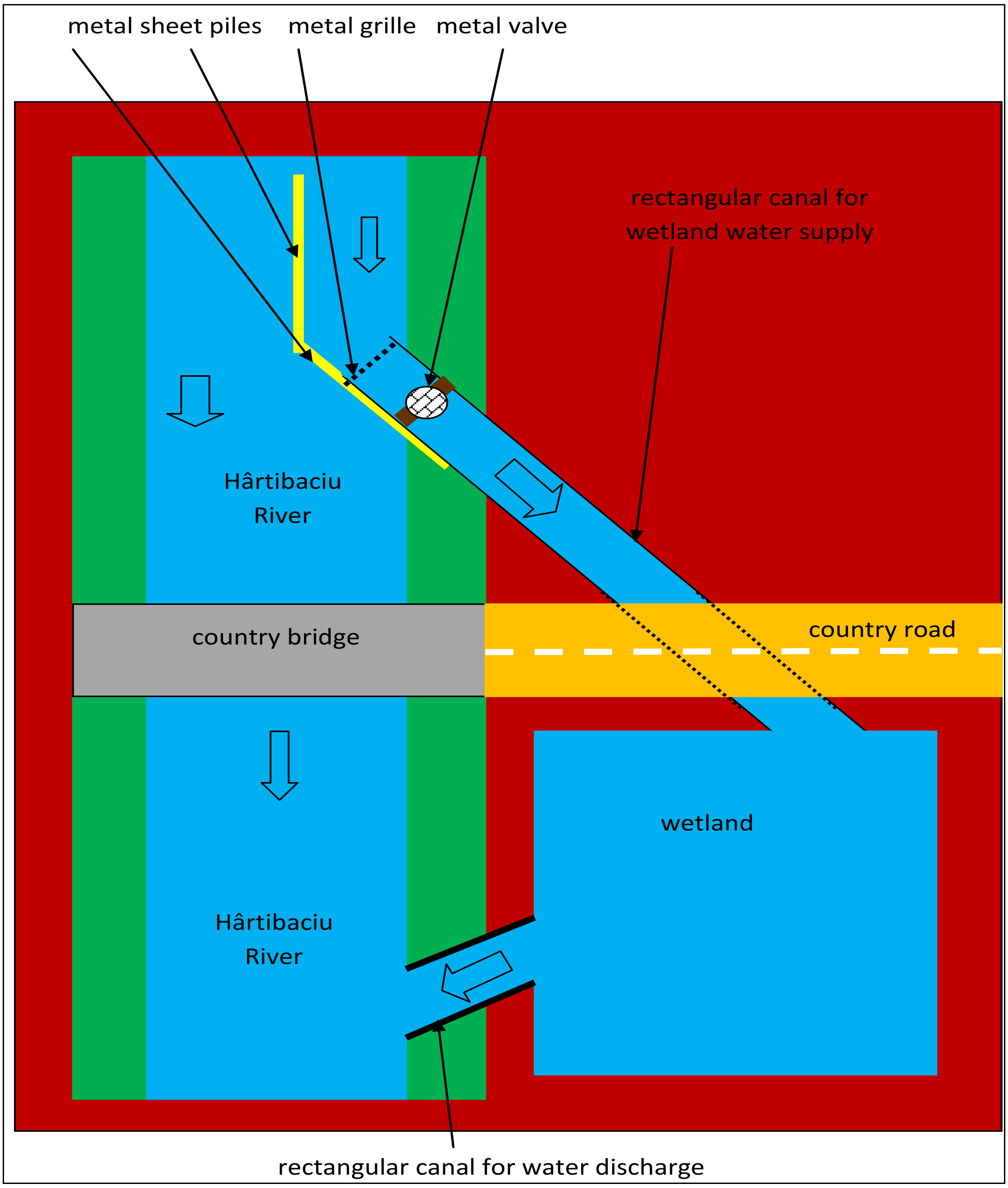


Figure 9: First water supply system of the wetland
– indicative scheme.

Second water supply system of the wetland

Another possibility regarding water supply system of the wetland after being arranged (Fig. 3b) is using the existing side canal (Fig. 10).

Figure 10: Existing side canal.

Thus, water abstraction from the Hârtibaciu River will be performed by the means of the same type of rectangular canal provided with some metal sheet pile at the upper end. (Fig. 4). Unlike the first water supply system described above, the abstracted water will not reach directly the wetland, but the side canal (the existing earth canal). Currently this canal is supplied by rainwater. Its arrangement is proposed in order to capture water from the Hârtibaciu River. Thus, its current shape will change into a trapezoidal one, and then the banks will be linearized (Fig. 11). The arranged canal should be without concrete, stones, etc., permitting by water movements the creation of small niches on the underground for species and as well permitting on the banks the planting of site typical willow species. These species offer on their roots valuable ecological niches for species.

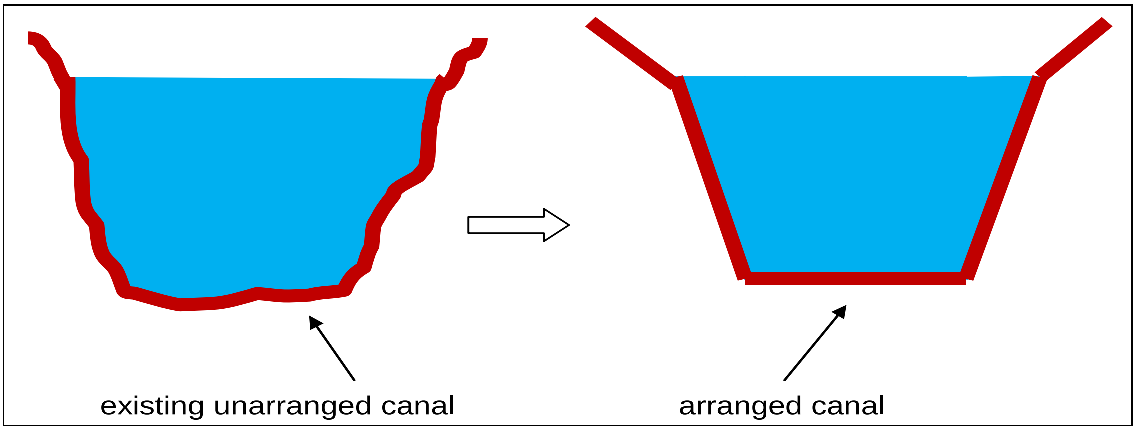


Figure 11: The arrangement of the side canal to supply the wetland.

For water abstraction a rectangular concrete basin upstream is to be built at the upstream end of this existing side canal. The basin will have the following dimensions: 3 m height, 1m length and 1.5 m width (Fig. 12).

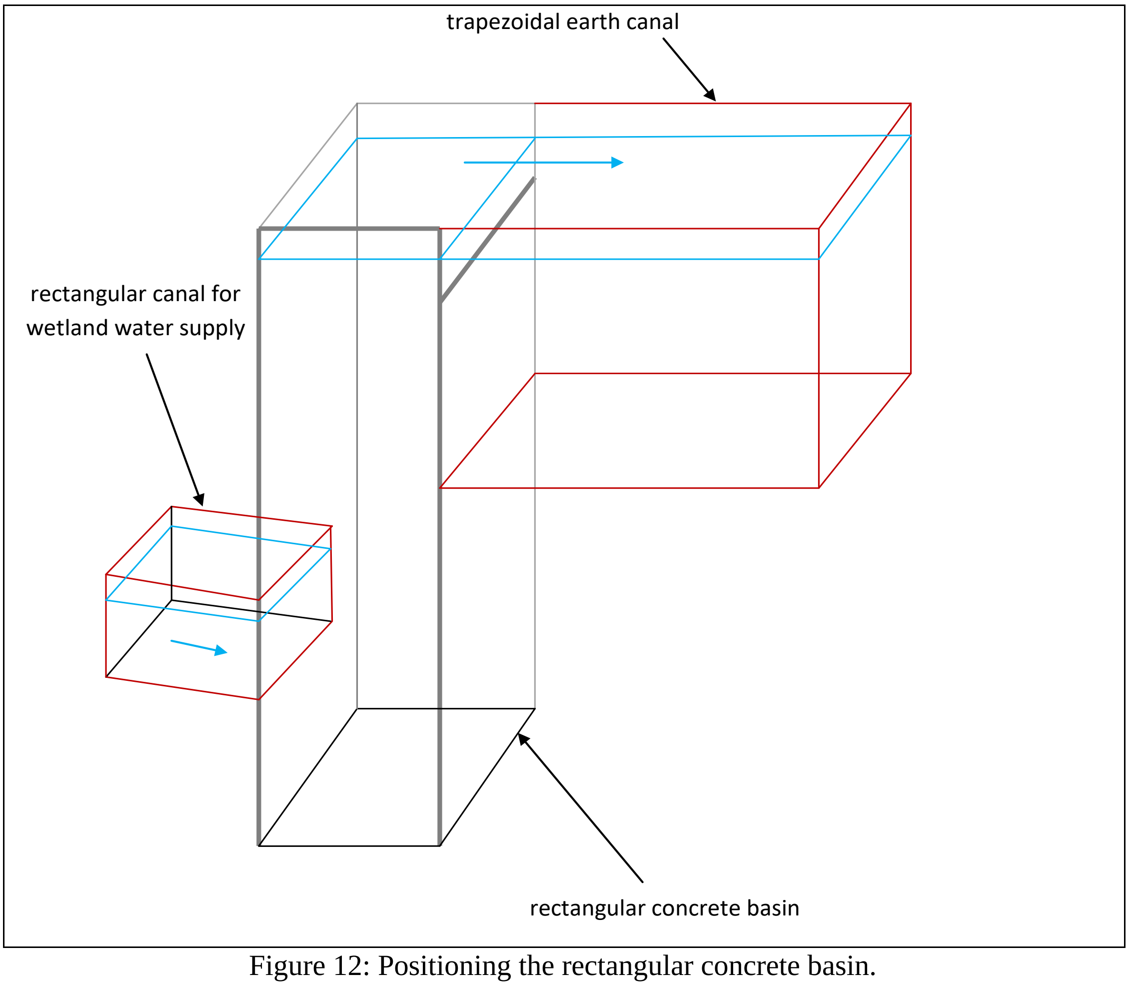


Figure 12: Positioning the rectangular concrete basin.

Throughout the trapezoidal canal length the wetland water supply will be achieved through four pipelines. At the end of the wetland, inside the trapezoidal canal, a sluice will be fixed (Fig. 13). The difference (waterfall) between the pipelines supplying the wetland gravitationally and the wetland water surface is of 1 meter. The flow of the four pipelines is the same with the one in the rectangular canal that abstracts water from the Hârtibaciu River.

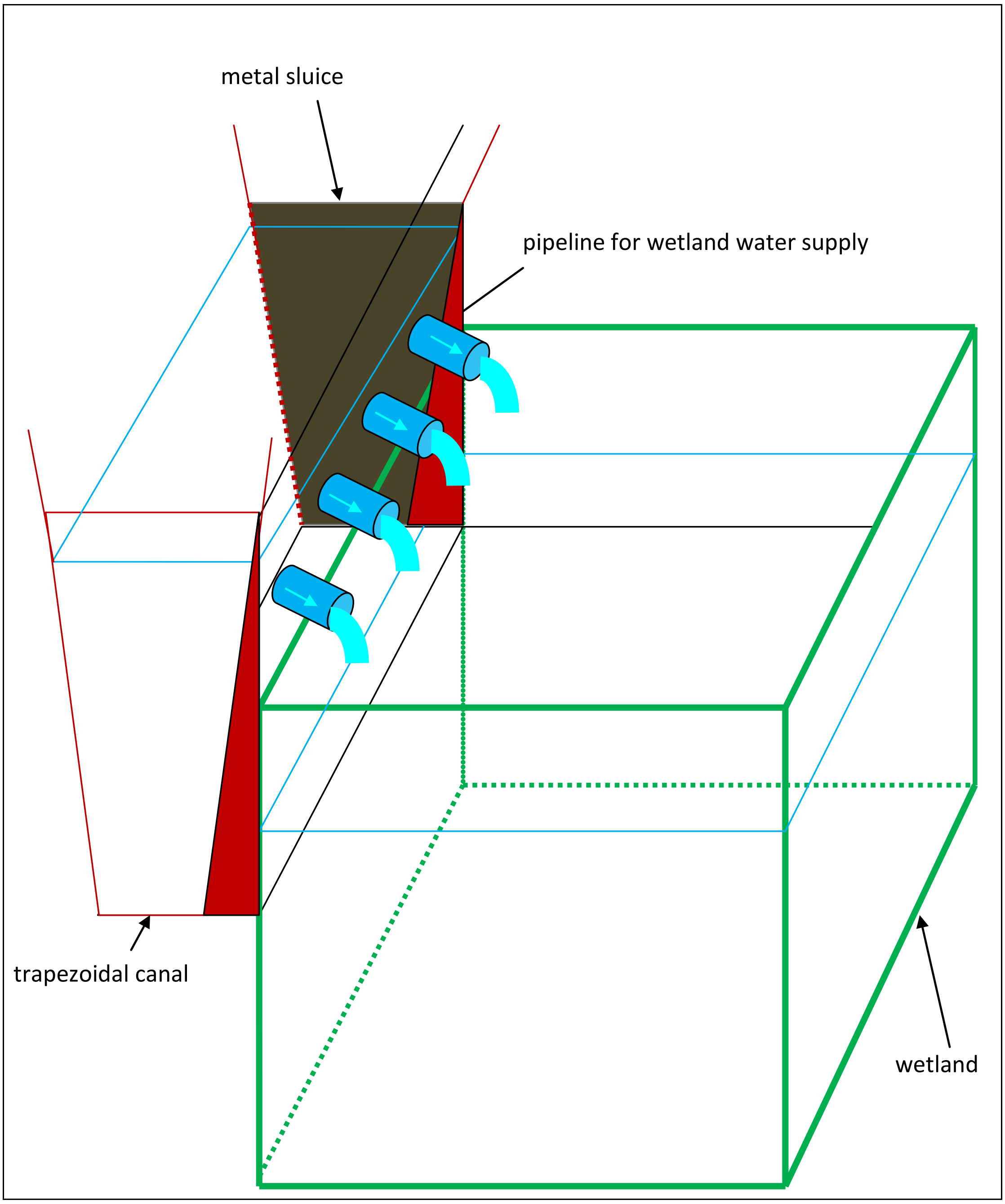


Figure 13: Positioning the water supply pipelines – indicative scheme.

Discharging water from the wetland (it is proposed that the water level in the wetland to be ~ 1m) into the Hârtibaciu River will be achieved through a rectangular canal having the same dimensions as the wetland supply canal (Fig. 14).

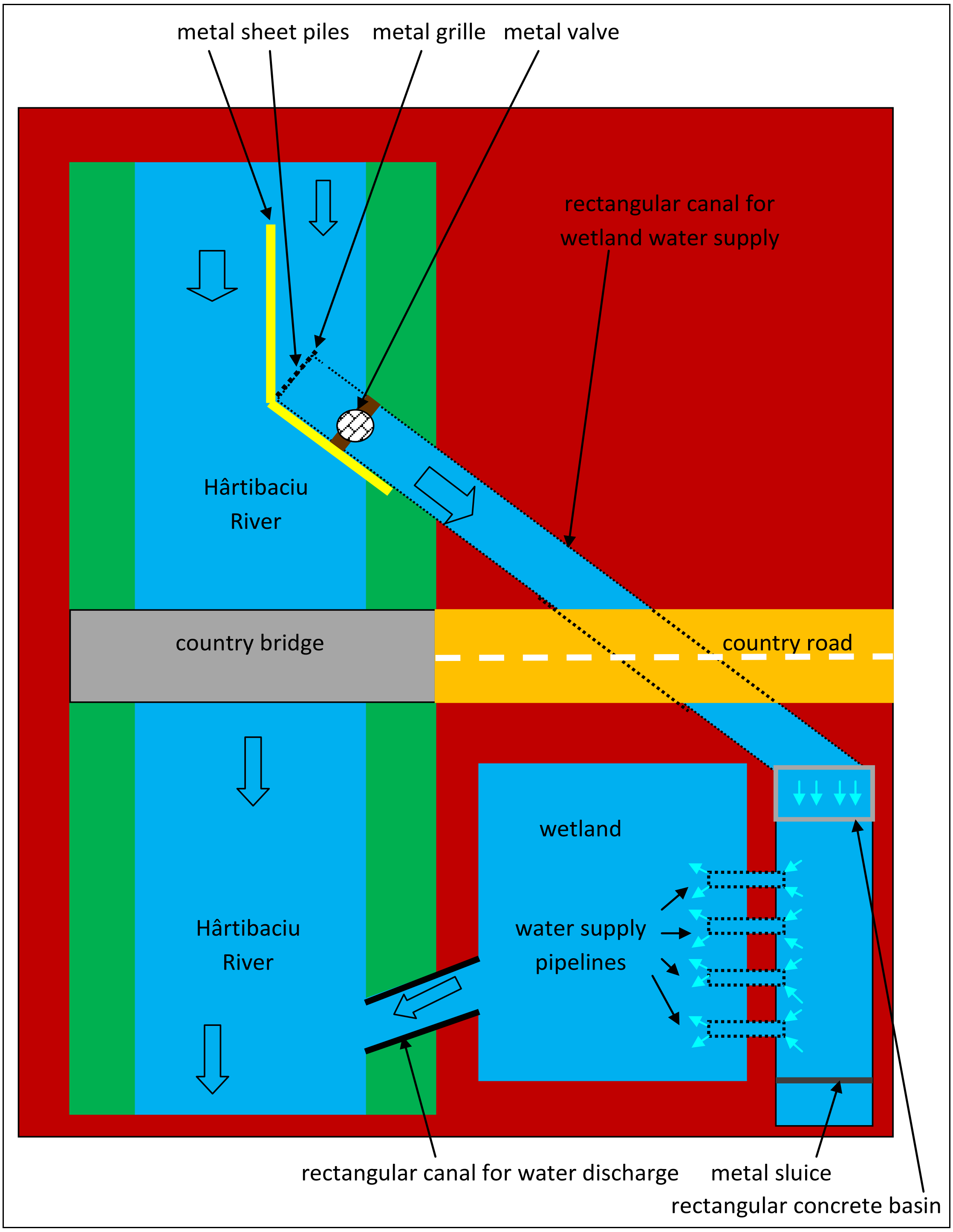


Figure 14: Second water supply system of the wetland – general indicative scheme.

CONCLUSIONS

The two important aspects of the solution proposed in this paper are:

1) The lateral connectivity improvement focusing on the gravitationally achieved water supply for the wetland and that the water discharge coming from the wetland will be directed gravitationally back into the Hârtibaciu River.

2) The local fish fauna will acquire new optimum flora enhanced habitats and the river system resilience will be increased in the studied lotic sector.

REFERENCES

1. Bănăduc D., Rey S., Trichkova T., Lenhardt M. and Curtean-Bănăduc A., 2016 – The Lower Danube River-Danube Delta-North West Black Sea: A pivotal area of major interest for the past, present and future of its fish fauna – A short review, *Science of the Total Environment*, 545-546, DOI: 10.1016/j.scitotenv.2015.12.05, 137-151.
2. Biswas A. K., 2008 – Integrated Water Resources Management: is it working?, *International Journal of Water Resources Development*, 24, 1, 5-22.
3. Bănărescu P. M. and Bănăduc D., 2007 – Habitats Directive (92/43/EEC) fish species (Osteichthyes) on the Romanian territory, *Acta Ichtiologica Romanica*, II, 43-78.
4. Curtean-Bănăduc A., Schneider-Binder E. and Bănăduc D., 2014 − The importance of the riverine ligneous vegetation for the Danube Basin lotic ecosystems, in Cianfaglione K. (ed.), L'importanza degli Alberi e del Bosco. Cultura, scienza e coscienza del territorio, Temi Ed., Trento, Italia, ISBN: 978-88-973772-63-9, I-II, 187-210.
5. Hapciuc O.-E., Romanescu G., Minea I., Iosub M., Enea A. and Sandu I., 2016 – Flood susceptibility analysis of the cultural heritage in the Suceviţa Catchment (Romania), *International Journal of Conservation Science*, 7, 2, ISSN: 2067-533X, 501-510.
6. Hoancă D., Todorescu C. and Roşu A., 2014 – Providing longitudinal connection in case of cross sluicing on water bodies in Banat Hydrographic area, *Transylvanian Review of Systematical and Ecological Research*, 16.2, 151-160, DOI: 10.1515/trser-2015-0024.
7. Jeeva V., Kumar S., Verma D. and Rumana H. S., 2011 – River fragmentation and connectivity problems in Gange River of upper Himalayas: the effect on the fish communities (India), *Transylvanian Review of Systematical and Ecological Research*, 12, The Wetlands Diversity, 75-90.
8. Kay E. L. and Voicu R., 2013 − Developing An Ecological And Migration System For Ichthyofauna On The Crişul Repede River Near The City Hall Of Oradea, *Management of Sustainable Developmen*, Sibiu, Romania, 5, 2, 27- 33.
9. Voicu R. and Liliana V., 2015 – The proposal of potential solutions in order to restore the Bârzeşti–Brăhăşoaia wetland within the Bârlad hydrographic basin, *Lakes, reservoirs and ponds*, 9, 2, 77-95
10. Voicu R. and Dominguez L., 2016 − Facilitation fish migration above the discharge sill located on the ialomiţa river near cave Ialomicioara, Annals of Valahia University of Targoviste, *Geographical Series*, 2016 16, 2, 44-58.
11. van Wilgen B. W., Le Maitre D. C. and Cowling R. M., 1998 – Ecosystem services, efficiency, sustainability and equity: working for water programme, *Trends in Ecology and Evolution*, 13, 9, 378.
12. Gumpinger C. and Scheder C., 2008 – Decline of biodiversity as a result of various human impacts related to river regulation – exemplified by several small river catchments (Austria), *Transylvanian Review of Systematical and Ecological Research*, 6, 141-148.
13. Ickes B. S., Vallazza J., Kalas J. and Knights B., 2005 − River floodplain connectivity and lateral fish passage: A literature review, U.S. Geological Survey, Upper Midwest Environmental Sciences Center, La Crosse, Wisconsin, June 2005, 25.

EFFECT OF VITAMIN E (DL-ALL-RAC-A-TOCOPHEROL ACETATE) AND NANO PARTICLES OF SELENIUM ON GROWTH, SURVIVAL, BODY COMPOSITION AND WHOLE BODY GLUTATHIONE PEROXIDASE (GPX) AND MALONDIALDEHYDE (MDA) IN CASPIAN KUTUM, *RUTILUS KUTUM* (KAMENSKY, 1901)

*Davoud TAHMASBI *, Saeid GORGIN **,*
Mohammad MAZENDARANI and *Mohammad SUDAGAR*

* Fisheries and Exploitation Department, College of Fisheries and Environment, Gorgan University of Agricultural Sciences and Natural Resources, Gorgan, Iran.
** Department of Fisheries and Exploitation, College of Fisheries and Environment, Gorgan University of Agricultural Sciences and Natural Resources, Gorgan, Iran, sgorgin@gau.ac.ir

DOI: 10.1515/trser-2017-0014

KEYWORDS: Vitamin E, nano-selenium, Growth performance, Glutathione peroxidase, Malondialdehyde.

ABSTRACT

The effect of vitamin E (100 mg kg^{-1}) and nano-selenium (1 mg kg^{-1}), which have a nutritional relationship separately and in combination, was investigated on growth, survival, carcass composition, body glutathione peroxidase activity, and body malondialdehyde content of Caspian kutum. Results showed that vitamin E is capable of improving growth, FCR and WG in Kutum fingerlings; however, nano-selenium is not. According to this study, vitamin E can improve growth and selenium can improve glutathione peroxidase activity in kutum larvae.

RESUMEN: Efecto de la vitamina E (DL aflta tocoferol acetato) y nano-partículas en el crecimiento, supervivencia, composición corporal, peroxidasa glutatión corporal (GPX) y malondialdehído en *Rutilus kutum* (Kamensky, 1901).

Se investigó el efecto de la vitamina E (100 mg kg^{-1}) y el nano-selenio (1 mg kg^{-1}), compuestos que poseen una relación nutricional tanto de forma separada como combinada, sobre el crecimiento, supervivencia, composición corporal, actividad del peroxidasa glutatión corporal y en el contenido de malondialdehído en el ciprínido de los Cárpatos *Rutilis kutum*. Los resultados mostraron que la vitamina E es capaz de mejorar el crecimiento en juveniles de la especie; sin embargo, el nano-selenio, no. Con acuerdo a este estudio, la vitamina E puede promover el crecimiento y el selenio puede aumentar la actividad de la peroxidasa glutatión en las larvas de *Rutilis kutum*.

REZUMAT: Efectul vitaminei E (DL-all-rac-α-tocoferol acetat) și a nanoparticulelor de seleniu asupra ratei de creștere, ratei de supraviețuire, compoziției corpului, activității glutation-peroxidazei (GPx) și concentrației generale de malondialdehidă în *Rutilus kutum* (Kamensky, 1901).

S-a studiat efectul vitaminei E (100 mg kg^{-1}) și nano-seleniului (1 mg kg^{-1}), cu rol nutrițional, administrate separate și combinat asupra ratei de creștere, ratei de supraviețuire, compoziției carcasei, asupra speciei *Rutilus kutum*. Rezultatele au arătat că vitamina E este capabilă să îmbunătățească creșterea, FCR și WG la puietul de *R. kutum*; însă, nano-seleniul nu a demonstrat efecte asupra acestor indicatori. Conform acestui studiu, vitamina E poate îmbunătăți creșterea iar seleniul poate îmbunătăți activitatea glutation-peroxidazei la alevinii de *R. kutum*.

INTRODUCTION

Caspian Kutum is one of the endemic species of the Caspian Sea, a popular fish, particularly in north Iran, due to meat quality and marketability (Ebrahimi et al., 2012; Afkhami et al., 2014). It is a valuable species for fishing industry (Ouraji et al., 2011) because of its distribution along the Caspian coastline (Heyrati et al., 2007). To restock the population of the Caspian Sea species the Iranian fisheries organization produces and releases a huge number of fingerlings annually, which, Caspian kutum comprises 89% of total bony fish species(Abdolhay et al., 2011). In restocking centers, Caspian Kutum larva feed on zooplanktons and a formulated diet (Ebrahimi et al., 2012). The formulated diet should be balanced with respect to all dietary components to allow the larvae to reach higher weight and to have adequate resistance to enter seawater.

Vitamin E is predicated to all materials acting as alpha-tocopherol (Parker and Parker, 2011). Tocopherols protect tissue against the damages caused by lipid oxidation (Hamre, 2011). Vitamin E is necessary to maintain meat quality, immune system, natural stability of blood cells against hemolysis, capillary permeability, and heart muscles (Halver and Hardy, 2002).

Selenium is one of the elements necessary in the natural life cycle of all organisms including fish (Köhrle, 2004). Selenium is important because it is involved in glutathione peroxidase (GPx) structure. This enzyme protects cells and tissues against oxidative damages (Watanabe et al., 1997).

Vitamin E and selenium have an obvious nutritional relationship. This relationship has been studied in a number of fish species. The results show that the symptoms of combined vitamin E and selenium shortage are muscular dystrophy, presence of muscular proteins in plasma and anemia (Bell et al., 1985; Bell et al., 1986; Bell et al., 1987; Gatlin et al., 1986; Poston et al., 1976).

Nano-materials have special properties. Recently, nano-selenium has attracted attention due to high bioavailability and low toxicity (Wang et al., 2007a). The aim of the present study was to investigate the singular and combined effects of vitamin E and nano-selenium on growth, survival, carcass composition, whole body GPx activity, and malondialdehyde content of Caspian kutum fingerlings. This was performed at the aquaculture laboratory of the Gorgan University of agricultural sciences and natural resources, Gorgan, Iran, over 6 weeks.

MATERIAL AND METHODS

Nano-selenium used in this study was provided from Nanosani Co. (Mashhad, Iran). Average particle size of this product was 50 nm. Also, Dl-all-rac-α-tocopherol acetate (Sigma Chemical Co., Steinheim, Germany) was used as a vitamin E source, because of high stability and bioavailability to fish (NRC, 1993). Basal diet composition is presented in table 1. Four diets were used in this study: 1 − the control diet without nano-selenium and vitamin E supplementation (control), 2 − the control diet supplemented with 100 mg/kg vitamin E (T-1), 3 − the control diet supplemented with 1 mg/kg nano-selenium (T-2), and 4 − the control diet supplemented with 100 mg/kg vitamin E and 1 mg/kg nano-selenium (T-3). To produce the diets dry ingredients were mixed and then oil and water were added to the mixture. Nano-selenium, being a solution, was added to the diets in combination with water, whereas Dl-all-rac-α-tocopherol acetate was added in combination with oil. The resultant dough was then passed through a mesh to form threads. The threads were air-dried and crushed into appropriate size before use.

Table 1: Formulation and proximate composition of experimental diets; * Vitamin mixture was manually provided according to feed requirements of the fish: (each kg^{-1} diet): vitamin A, 10,000 IU; vitamin D_3 2000 IU; vitamin K, 20 mg; vitamin B_1, 400 mg; vitamin B_2, 40 mg; vitamin B_6, 20 mg; vitamin B_{12}, 0.04 mg; choline chloride, 1200 mg; folic acid, 10 mg; niacin, 200 mg; ** Mineral mixture (mg g^{-1}) : $CaCO_3$ 36; KH_2PO_4 502; $MgSO_4.7H_2O$ 162.

Ingredients	%
Soybean meal	34
Fish meal	30
Corn flour	13
Wheat flour	12
Fish oil	7
Vitamin mix*	2
Mineral mix**	2
Proximate composition	%
Moisture	11.3
Crude protein	38.8
Lipid	8.4
Ash	10.5

Fish and experimental conditions

Caspian kutum fingerlings with the average weight of 250 ± 0.03 mg, were provided from Teleost Propagation Center of Sijaval, Bandar Torkman, Iran. The fish were allowed to acclimatize to experimental condition in two 500 l tanks for 2 weeks. During the acclimation period the fish were fed with commercial feed (kutum starter feed, protein = 30%, fat = 10%; Mazandaran Feed Co.). After the acclimation, 480 fish were stocked into 12 tanks (100 l). Each of the aforementioned diets were offered to three tanks.

During the trial, fish were fed with their corresponding diet (12% body weight per day) thrice a day (08:00, 12:00 and 16:00). Wastes were daily siphoned from the tanks. Fifty percent of the tanks' water content was replaced with fresh water daily. Water quality was periodically monitored. Temperature was 20.3-24.8, pH was 7.3-7.6 and oxygen was higher than 7.8 ppm. The fish were maintained under natural photoperiod during the experiment.

The diets and fish carcass composition was determined by standard methods suggested by (AOAC, 2002). Moisture content was determined with oven at 105ºC until reaching to a constant weight. Ash was determined by combustion at 550ºC for 12 h. Protein was determined by Kjeldahl method after acid digestion. Fat was determined by Soxhelt apparatus using ether petroleum for 6 h. The fish weight was recorded at the experiment initiation and fortnightly thereafter to adjust feed amount. The feed conversion ratio (FCR) and weight gain (WG%) were evaluated by the following formulas:Weight gain (WG%) = $[(W_F-W_I)/W_I]\times100$
Feed conversion ratio (FCR) = [Total feed intake(g) / Total wet weight gain (g)].

To determine GPx and MDA, the fish whole body was first washed in sterile phosphate buffered saline (137 mM NaCl, 2.7mM KCl, 4.3 mM $Na_2HPO_4.7H_2O$, pH 7.3) and then 1 g sample was homogenized in 10 ml in same buffer. The homogenized sample was centrifuged (5000 g) and supernatant was separated for assay. The supernatant was stored at -70 ºC until analyses.

GPx was determined according to Bell et al. (1986) and expressed as U mg^{-1} protein. The supernatant total protein was determined according to Peterson (1977). Bovine serum albumin was used as standard. The sample MDA was determined by Kei (1978) method.

Statistics were performed using SPSS program version 16. One way ANOVA was used to determine the significant ($P < 0.05$) difference among the groups.

RESULTS AND DISSCUSION

Growth and carcass composition

T-1 and T-3 groups had a significantly ($P < 0.05$) higher final weight and weight gain, as well as lower FCR, compared to the control and T-2 groups (Tab. 2). There was no significant ($P > 0.05$) difference in survival among the treatments (Tab. 2). There was no significant difference ($P > 0.05$) in carcass moisture, protein, lipid, and ash content among the treatments (Tab. 3).

Table 2: Effect of vitamin E and nano-selenium of growth parameters of Caspian Kuttum.

Group/treatment	control	T-1	T-2	T-3
Initial weight (g)	$0.27±0.02^a$	$0.23±0.02^a$	$0.26±0.03^a$	$0.22±0.04^a$
Final weight (g)	$0.86±0.02^b$	$0.94±0.01^a$	$0.85±0.02^b$	$0.92±0.03^a$
Survival rate (%)	$95±2.5^a$	$96.6±1.4^a$	95^a	$97.5±2.5^a$
WG	$212±21^b$	$299±37^a$	$223±26^b$	$321±61^a$
FCR	$4.2±0.2^b$	$3.6±0.2^a$	$4.3±0.2^b$	$3.6±0.2^a$

Table 3: Whole-body proximate compositions (% live weight basis) of kutum fingerlings after feeding trial.

Composition	Control	T-1	T-2	T-3
Moisture	$73.1± 0.4^a$	$73.6± 0.6^a$	$73.5± 0.6^a$	$73±0.9^a$
Crude protein	$12.4± 0.3^a$	$12.5± 0.2^a$	$12.5± 0.2^a$	$12.7± 0.1^a$
Crude lipid	$11.2± 0.4^a$	$10.9± 0.3^a$	$11.4± 0.3^a$	$11.1± 0.2^a$
Ash	$2.4± 0.2^a$	$2.4± 0.1^a$	$2.3± 0.2^a$	$2.5± 0.1^a$

Whole body GPx and MDA

GPx activity of the control and T-1 groups were similar and significantly ($P < 0.05$) lower than T-2 and T-3 groups (Fig. 1). There was no significant difference ($P > 0.05$) in GPx activity between T-2 and T-3 groups (Fig. 1). There was no significant difference ($P > 0.05$) in MDA content among the treatments (Fig. 2).

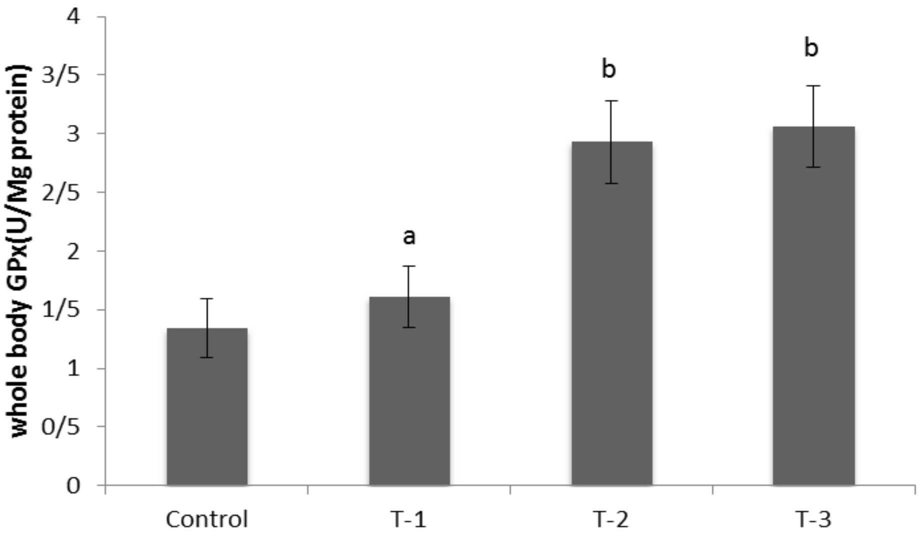


Figure 1: Effect of vitamin E and nano-selenium on body GPx activity of Caspian Kutum.

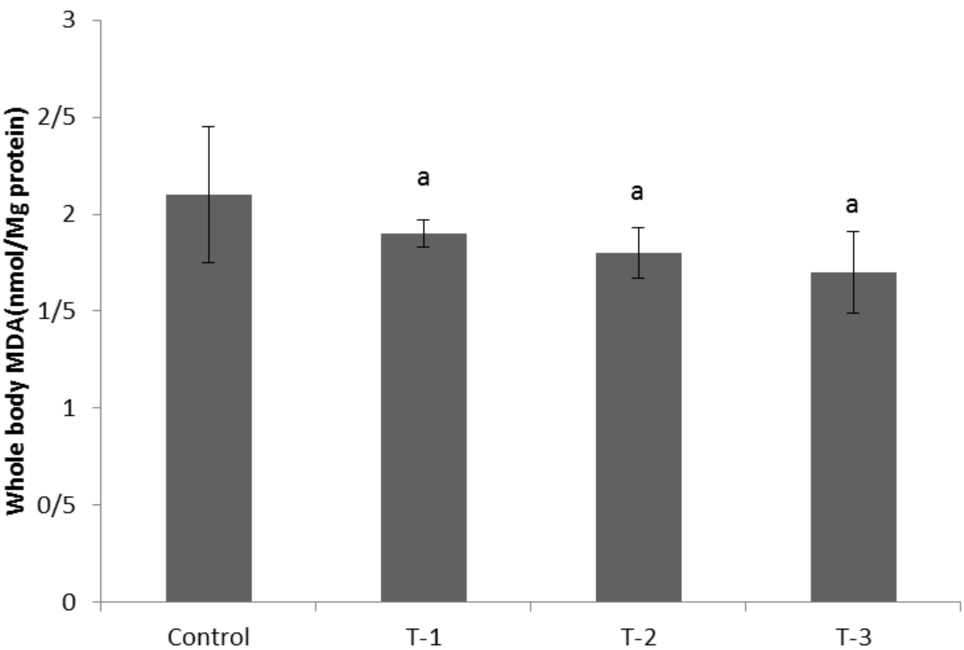


Figure 2: Effect of vitamin E and nano-selenium on whole body MDA content of Caspian Kutum.

DISCUSSION

The results of this study showed that the treatments containing vitamin E were significantly different from other treatments with respect to final weight, FCR and WG. Thus it can be stated that vitamin E was the factor responsible for growth promotion, furthermore, previous studies revealed the effect of vitamin E on growth promotion (Huang and Huang, 2004; Peng et al., 2009; Amlashi et al., 2011). According to the results obtained in the present study, it can be expressed that nano-selenium had no effect on growth, FCR and GW of Caspian Kutum. This result is not in line with Zhou et al. (2009), which showed that nano-selenium improved growth and FCR in *Carassius auratus gibelio*. The reasons for this contradiction could be due to lower FCR in Caspian Kutum compared to *C. auratus gibelio*, the presence of selenium in the diets' ingredients, and presence of vitamin E or other antioxidants, such as vitamin C, in the diets covering the role of selenium. However, feeding Senegalese sole (Solea senegalensis) larvae on live food supplemented with organic selenium (Sel-Plex containing selenomethionin) had no beneficial effects on growth and survival (Ribeiro et al., 2012). Also, in another study on *Oncorhynchus mykiss*, growth was not affected by different levels of Se-yeast and selenite (Rider et al., 2009), which is in line with the present study.

Selenium is a structural component of glutathione peroxidase (Rotruck et al., 1973). In the present study, the whole body GPx activity was measured. The fish receiving the diet containing nano-selenium had higher GPx activity than the other fish, confirming the effect of nano-selenium on GPx activity. Previous studies on the effect of nano-selenium (Zhou et al., 2009) and inorganic or organic selenium (Wang et al., 2007b; Ribeiro et al., 2012; Bell et al., 1985) on fish showed that plasma or liver GPx activity increased because of selenium supplementation. In the present study, GPx activity of the control and T-1 groups were approximately similar. Therefore, it can be stated that vitamin E, without selenium presence, cannot modulate GPx activity. Previous studies on rainbow trout (Bell et al., 1985) and channel catfish (Wise et al., 1993) showed similar results. MDA, produced as a result of lipid peroxidation, is measured as oxidative stress indicator (Esterbauer et al., 1991). In the present study, whole body MDA among the treatments was not significantly different, which could be due to the presence of other antioxidants such as vitamin C in the diets or lack of oxidative conditions during the experiment. Similar to the present study, Zhu et al. (2012) reported an increase in liver GPx activity and no change in MDA content in *Micropterus salmoides* as a result of feeding on selenium-supplemented diet. Oxidative stress can be induced by a large variety of conditions, including nutritional imbalance, exposure to chemical and physical agents in the environment, strenuous physical activities, injury, and hereditary disorders (Chow, 1991). The Caspian Sea is polluted with heavy metals, oil, industrial wastewater, etc. These pollutions can induced oxidative stress and increased mortality at the time of entering the kutums larva into the sea. According to previous studies (Özkan-Yılmaz et al., 2013; Mittler, 2002), GPx by decreasing free radicals and finally MDA can increase the resistance of fish against the oxidative stress. On the other hand presence of vitamin E on diet can improve the final weight, that increase in the final weight of fish can improve the resistance of fish against unsuitable conditions. Overall, the results of the present study showed that nano-selenium could not affect growth performance without the presence of vitamin E. Likewise, vitamin E failed to increase GPx activity, without the presence of selenium. Therefore, it is concluded that both nano-selenium and vitamin E should be added to Caspian Kutum diet to increase growth performance and antioxidant enzyme, particularly GPx, activity. Such conditions can lead to increase in resistance of Caspian Kutum fingerling while entering the sea and afterwards.

REFERENCES

1. Abdolhay H. A., Daud S. K., Ghilkolahi S. R., Pourkazemi M., Siraj S. S. and Satar M. A., 2011 − Fingerling production and stock enhancement of Mahisefid (*Rutilus frisii kutum*) lessons for others in the south of Caspian Sea, *Reviews in Fish Biology and Fisheries,* 21, 247-257.
2. Afkhami M., Bastami K. D., Shabani N. and Soltani F., 2014 – A survey on ionic and metabolite factors of blood serum in Kutum (Rutilus frisii kutum), *Transylvanian Review of Systematical and Ecological Research*, 16.2, The Wetlands Diversity, 127-132.
3. Amlashi A. S., Falahatkar B., Sattari M. and Gilani M., 2011 − Effect of dietary vitamin E on growth, muscle composition, hematological and immunological parameters of sub-yearling beluga (Huso huso) L., *Fish and shellfish immunology,* 30, 807-814.
4. AOAC, 2002 − Official Methods of Analysis of AOAC International (17th edn), Arlington, VA.
5. Bell J., Cowey C., Adron J. and Pirie B.,1987 − Some effects of selenium deficiency on enzyme activities and indices of tissue peroxidation in Atlantic salmon parr (Salmo salar), *Aquaculture,* 65, 43-54.
6. Bell J., Cowey C., Adron J. and Shanks A. M., 1985 − Some effects of vitamin E and selenium deprivation on tissue enzyme levels and indices of tissue peroxidation in rainbow trout (Salmo gairdneri), *British Journal of Nutrition,* 53, 149-157.
7. Bell J., Pirie B., Adron J. and Cowey C., 1986 − Some effects of selenium deficiency on glutathione peroxidase (EC 1.11. 1.9) activity and tissue pathology in rainbow trout (Salmo gairdneri), *British Journal of Nutrition,* 55, 305-311.
8. Chow C. K., 1991 − Vitamin E and oxidative stress, *Free Radical Biology and Medicine,* 11, 215-232.
9. Ebrahimi G., Ouraji H., Firouzbakhsh F. and Makhdomi C., 2012 − Effect of dietary lipid and protein levels with different protein to energy ratios on growth performance, feed utilization and body composition of Rutilus frisii kutum (Kamenskii, 1901) fingerlings, Aquaculture Research.
10. Esterbauer H., Schaur R. J. and Zollner H., 1991 − Chemistry and biochemistry of 4-hydroxynonenal, malonaldehyde and related aldehydes, *Free Radical Biology and Medicine,* 11, 81-128.
11. Gatlin D., Poe W. E. and Wilson R. P.,1986 − Effects of singular and combined dietary deficiencies of selenium and vitamin E on fingerling channel catfish (Ictalurus punctatus), *The Journal of nutrition,* 116, 1061-1067.
12. Halver J. E. and Hardy R. W., 2002 − Nutrient flow and retention, *Fish nutrition,* 3, 755-770.
13. Hamre K., 2011 − Metabolism, interactions, requirements and functions of vitamin E in fish, *Aquaculture Nutrition,* 17, 98-115.
14. Heyrati F. P., Mostafavi H., Toloee H. and Dorafshan S., 2007 − Induced spawning of kutum, Rutilus frisii kutum (Kamenskii, 1901) using (D-Ala 6, Pro 9-NEt) GnRHa combined with domperidone, *Aquaculture,* 265, 288-293.
15. Huang C.-H. and Huang S.-L.., 2004 − Effect of dietary vitamin E on growth, tissue lipid peroxidation, and liver glutathione level of juvenile hybrid tilapia, (Oreochromis niloticus)×(O. aureus), fed oxidized oil, *Aquaculture,* 237, 381-389.
16. Kei S., 1978 − Serum lipid peroxide in cerebrovascular disorders determined by a new colorimetric method, *Clinica Chimica Acta,* 90, 37-43.
17. Köhrle J., 2004 − Selenium in biology and medicine − further progress and increasing interest, *Journal of Trace Elements in Medicine and Biology,* 18, 61-63.
18. Mittler R., 2002 − Oxidative stress, antioxidants and stress tolerance, *Trends in plant science,* 7, 405-410.
19. NRC., 1993 − Nutrient requirements of fish (NRC), *Washington, DC: National Academic Press*.
20. Ouraji H., Khalili K. J., Ebrahimi G.and Jafarpour S. A., 2011 − Determination of the optimum transfer time of kutum (Rutilus frisii kutum) larvae from live food to artificial dry feed, *Aquaculture International,* 19, 683-691.

21 Özkan-Yılmaz F., Özlüer-Hunt A., Gündüz S. G., Berköz M. and Yalın S., 2013 − Effects of dietary selenium of organic form against lead toxicity on the antioxidant system in Cyprinus carpio, *Fish physiology and biochemistry*, 1-9.
22. Parker R. and Parker R., 2011 − Aquaculture science, CengageBrain. com.
23. Peng S., Chen L., Qin J., Hou J., Yu N., Long Z., Li E. and Ye J., 2009 − Effects of dietary vitamin E supplementation on growth performance, lipid peroxidation and tissue fatty acid composition of black sea bream (Acanthopagrus schlegeli) fed oxidized fish oil, *Aquaculture Nutrition*, 15, 329-337.
24. Peterson G. L.,1977 − A simplification of the protein assay method of Lowry et al. which is more generally applicable, *Analytical biochemistry*, 83, 346-356.
25. Poston H. A., Combs Jr. G. F. and Leibovitz L., 1976 − Vitamin E and selenium interrelations in the diet of Atlantic salmon (Salmo salar): gross, histological and biochemical deficiency signs, *The Journal of nutrition*, 106, 892.
26. Ribeiro A., Ribeiro L., Sæle Ø., Hamre K., Dinis M. and Moren M., 2012 − Selenium supplementation changes glutathione peroxidase activity and thyroid hormone production in Senegalese sole (Solea senegalensis) larvae, *Aquaculture Nutrition*, 18, 559-567.
27. Rider S. A., Davies S. J., Jha A. N., Fisher A. A., Knight J. and Sweetman J. W., 2009 − Supra-nutritional dietary intake of selenite and selenium yeast in normal and stressed rainbow trout (Oncorhynchus mykiss): Implications on selenium status and health responses, *Aquaculture*, 295, 282-291.
28. Rotruck J., Pope A., Ganther H., Swanson A., Hafeman D. G. and Hoekstra W., 1973 − Selenium: biochemical role as a component of glutathione peroxidase, *Science*, 179, 588-590.
29. Wang H., Zhang J. and Yu H., 2007a − Elemental selenium at nano size possesses lower toxicity without compromising the fundamental effect on selenoenzymes: comparison with selenomethionine in mice, *Free Radical Biology and Medicine*, 42, 1524-1533.
30. Wang Y., Han J., Li W. and Xu Z., 2007b − Effect of different selenium source on growth performances, glutathione peroxidase activities, muscle composition and selenium concentration of allogynogenetic crucian carp (Carassius auratus gibelio), *Animal feed science and technology*, 134, 243-251.
31. Watanabe T., Kiron V. and Satoh S., 1997 − Trace minerals in fish nutrition, *Aquaculture*, 151, 185-207.
32 Wise D., Tomasso J., Gatlin III D., Bai S. and Blazer V., 199? − Effects of dietary selenium and vitamin E on red blood cell peroxidation, glutathione peroxidase activity, and macrophage superoxide anion production in channel catfish, *Journal of Aquatic Animal Health*, 5, 177-182.
33. Zhou X., Wang Y., Gu Q. and Li W., 2009 − Effects of different dietary selenium sources (selenium nanoparticle and selenomethionine) on growth performance, muscle composition and glutathione peroxidase enzyme activity of crucian carp (Carassius auratus gibelio), *Aquaculture*, 291, 78-81.
34. Zhu Y., Chen Y., Liu Y., Yang H., Liang G. and Tian L., 2012 − Effect of dietary selenium level on growth performance, body composition and hepatic glutathione peroxidase activities of largemouth bass Micropterus salmoide, *Aquaculture Research*, 43, 1660-1668.

THE INVASIVE RACCOON DOG (*NYCTEREUTES PROCYONOIDES*, GRAY) – AN UPDATE OF ITS DISTRIBUTION ON THE BALKANS

Elitsa POPOVA * and *Diana ZLATANOVA* *

* Sofia University "St. Kliment Ohridski", Faculty of Biology, Department of Zoology and Anthropology, Dragan Tsankov Boulevard 8, Sofia, Bulgaria, BG-1164, elitsa.d.popova@gmail.com, zlite2@gmail.com

DOI: 10.1515/trser-2017-0015

KEYWORDS: mining impact, lenitic ecosystems, lotic ecosystems, fish, macroinvertebrates, pressures, threats, risk management.

ABSTRACT

The raccoon dog, which lives especially near water and is rare in areas with low humidity, has been spreading throughout Europe since its introduction to Western Russia in the beginning of the 20th century. Official accounts of its distribution in Europe are often inaccurate due to scarce data. A literature search was conducted to identify records of the raccoon dog in the Balkans. More than 60 records were identified, including ones unlisted by the cited source from central and western Bulgaria, southern Serbia, Macedonia, Bosnia, Herzegovina, and Greece. The raccoon dog can be found on the Balkans either along the Danube (which is a major corridor for its invasion) or along its tributaries, which might represent secondary invasion pathways.

ZUSAMMENFASSUNG: Der invasive Marderhund *Nyctereutes procyonoides* Gray - eine Aktualisierung seiner Verbreitung auf dem Balkan.

Der Marderhund der besonders in der Nähe von Wasser lebt und in Gebieten mit geringer Feuchtigkeit selten ist, hat sich seit seiner Einführung im Westen Russlands Anfang des 20. Jahrhunderts in ganz Europa ausgebreitet. Offizielle Zahlen für seine Ausbreitung in Europa sind aufgrund knapper Daten oft ungenau. Um Aufzeichnungen über das Vorkommen des Marderhundes auf dem Balkan zu finden, wurde eine Literaturrecherche durchgeführt. Dabei wurden mehr als 60 Einträge identifiziert, darunter solche, die in der zitierten Quelle aus Zentral- und Westbulgarien, Südserbien, Mazedonien, Bosnien und Herzegowina und Griechenland nicht gelistet wurden. Die meisten Fundorte des Marderhunds auf dem Balkan liegen entweder entlang der Donau (die ein wichtiger Wildkorridor für seine Invasion ist) oder entlang ihrer Nebenflüsse, die sekundäre Invasionswege darstellen könnten.

REZUMAT: Specia invazivă câinele enot (*Nyctereutes procyonoides*, Gray) – o aducere la zi a distribuţiei sale în Balcani.

Câinele enot, care trăiește mai ales în apropierea apei și este rar în zonele cu umiditate scăzută, s-a răspândit în toată Europa, de la introducerea sa în vestul Rusiei la începutul secolului al 20-lea. Semnalările oficiale ale distribuției sale în Europa sunt adesea inexacte, din cauza datelor insuficiente. S-a efectuat o căutare în literatura de specialitate pentru a identifica înregistrările câinelui enot din Balcani. Au fost identificate mai mult de 60 de înregistrări, inclusiv cele din surse nelistate, citate din Bulgaria centrală și de vest, sudul Serbiei, Macedoniei, Bosniei și Herțegovina și Greciei. Cele mai multe dintre locurile cu câini enot din Balcani sunt fie de-a lungul Dunării (care este un coridor major pentru invazie) fie de-a lungul afluenților săi, care ar putea reprezenta căi secundare de invazie.

INTRODUCTION

The raccoon dog (*Nyctereutes procyonoides*, Gray) is native to East Asia, but it has been introduced to the Western parts of modern day Russia in the first half of the 20th century. Since then, its range has been expanding throughout Europe, reaching Finland and Sweden to the North, France and Germany to the West, and Romania and Bulgaria to the South (Kauhala and Kowalczyk, 2011; Kauhala and Saeki, 2016).

It has become one of the most successful alien carnivores in Europe, due to its adaptability, omnivory, and high reproductive potential (Kauhala and Kowalczyk, 2011). Its effects on the local wildlife are still poorly understood in many countries.

Data for the raccoon dog's distribution on the Balkans is scarce,which might have caused inaccuracies in the current distribution maps (Kauhala and Kowalczyk, 2011; Kauhala and Saeki, 2016).

The aim of this study was to collect new or omitted records for raccoon dog presence from the Balkan countries and to provide data for an update of the distribution range. This is vital when studying the species' invasion corridors and its potential effects, and could guide future management actions.

Figure 1: *Nyctereutes procyonoides*, Gray; photo: Kuczynski P.

MATERIAL AND METHODS

There are slight differences between the two raccoon dog maps presented by Kauhala and Kowalczyk, 2011 and the IUCN Red List of Threatened Species (Kauhala and Saeki, 2016), despite the fact that they have a common author. It is unclear how these maps were created and whether the differences are due to spatial inaccuracies or an update of the latter source. Regarding the Balkans, the discrepancies are minimal. For simplicity, we consider only the map accepted by IUCN. We conducted a literature search based on the key words "raccoon dog" and "*Nyctereutes procyonoides*" in combination with the names of the Balkan countries (Bulgaria, Serbia, Macedonia, Turkey, Croatia, Bosnia, and Herzegovina in the web search engine Google Scholar. The racoon dog is reported to inhabit the whole country of Romania, so no search was conducted for it). An additional search was conducted in Google to detect grey literature on the subject, including management plans for protected areas and online forums for hunters. All of the identified records were classified in 2 categories (following the methodology applied by Cirovic and Milenkovic, 1999): reliable evidence (captured individuals, photographs, tracks or others, reported by experts) and records without reliable evidence (observations or killed individuals, reported by non-experts) according to the way they were documented. ArcGIS v.10 (ESRI, 2011) was used to map the locations, together with the current distribution map.

RESULTS AND DISCUSSION

A total of 62 new records were identified in six Balkan countries (Tab. 1), of which 42% (n = 26) were records with reliable proof and 58% (n = 36) were without reliable proof. Most of the locations lay outside the current distribution range. We suggest that the Southern border of the range should be moved further south to include at least the reliable records in Northern Bulgaria and Eastern and Northwestern Serbia. These are close to the areas inhabited by the raccoon dog and are likely to be occupied.

Table 1: List of the newly identified locations of raccoon dogs on the Balkans.

Country	No. of locations		Total	Time range	Sources
	with reliable evidence	without reliable evidence			
Bosnia and Herzegovina*	-	1	1	1987	Cirovic and Milenkovic, 1999;
Bulgaria	13	21	34	1967-2015	Dragoev, 1978; Genov, 2012; Georgiev, 2010; Green Balkans, 2001; Natchev, 2016; Peshev and Yordanov, 1968; management plans (in project) for NP "Bulgarka", NP "Rusenski Lom" and MR "Atanasovsko ezero";
Croatia	1	1	2	1994-2016	Cirovic and Milenkovic, 1999; Duplić et al., 2016;
Greece	2	1	3	2005-2009	Adamopoulou and Legakis, 2016; Catsadorakis and Bousbouras, 2010;
Macedonia	1	-	1	2001	Ćirović, 2006;
Serbia*	10	12	22	1978-2015	Cirovic and Milenkovic, 1999; Milačič et al., 2015;

* Some of these locations are recorded during the time these countries were part of Federal Republic of Yugoslavia.

The only reliable record from Southern Bulgaria is a camera trap photo taken in the mountainous area of Shiroka Polyana dam (around 1500 m.a.s.l) which is surprising, since the most preferred habitats by the species in Europe are at much lower altitudes (Barrat et al., 2010; Drygala et al., 2008; Kauhala and Saeki, 2004). The reliable records in Greece, Macedonia and Northwestern Croatia are isolated and might represent cases of dispersing individuals rather than established populations. However, they may indicate further invasion towards the south and west on the Balkans than previously taught.

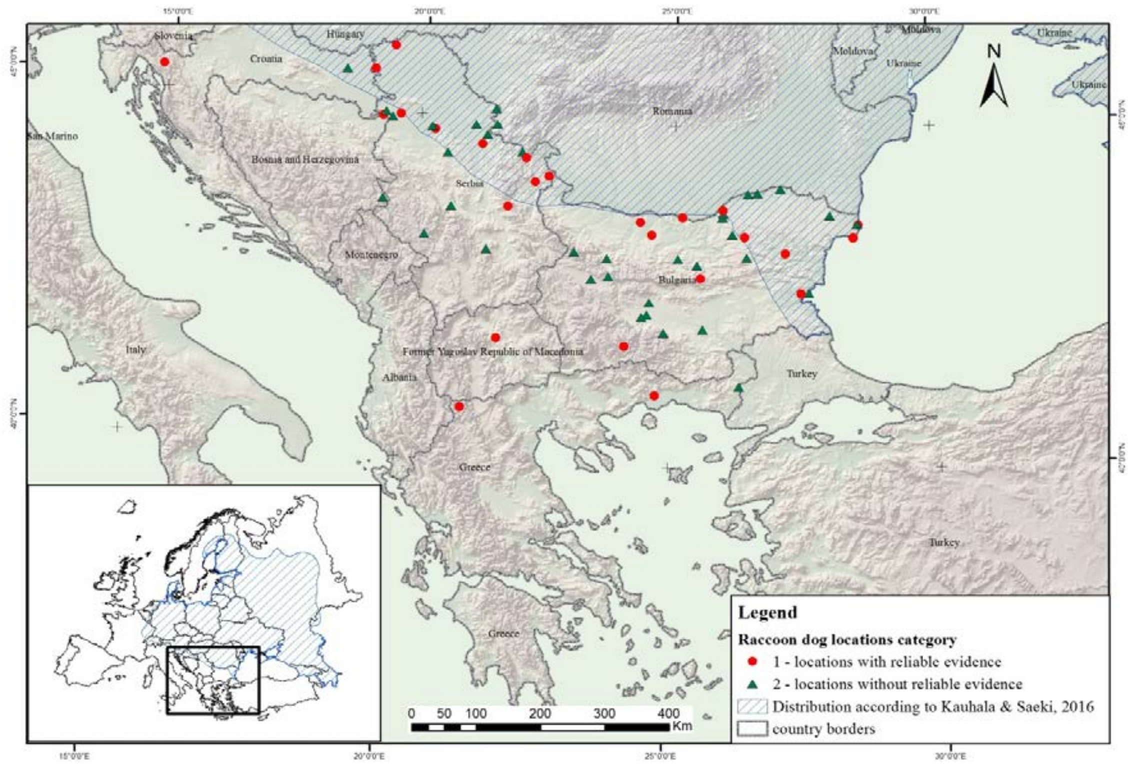


Figure 1: Distribution of the raccoon dogs on the Balkans – distribution and new records.

Many of the collected records lack exact coordinates and only a few contain information regarding habitat characteristics. However, we were able to identify the following places visited by raccoon dogs (only those marked with * are reliable records): lake shores (Bulgaria: Shabla* and Srebarna lakes, Greece: Prespa lake*), river shores (Bulgaria: Danube*, Chaya river; Greece – Nestos (Mesta) river*; Serbia: Danube*, Sava river*, Boljetinska river), Danube island (Bulgaria: Belene island*), Black sea shore (Bulgaria: Bolata cove*), marshes (Bulgaria: Kalimok marsh), agricultural airport (Bulgaria: Krasen village), landfills (Bulgaria: General Toshevo village), brines (Bulgaria: Pomorie lake), dams (Bulgaria: Pyasachnik dam), rice paddies (Bulgaria: near Plovdiv) and garden at the outskirts of a village (Bulgaria: Malo Konare village). In two cases raccoon dogs were found on a road, run over by a car (Macedonia: halfway from Titov Veles to Katlanovo*; Greece: by Dialekto, Xanthi*). All of the other records report only the general location (e.g. “surroundings of Nepotin”). Most of the reliable records are along the Danube, which is an established invasion corridor for the raccoon dog. Many of the others are also near bodies of water (smaller rivers or lakes), which is in accordance with the known habitat preferences of the species (Barrat et al., 2010; Kauhala and Saeki, 2004).

It is evident that most of the data regarding the raccoon dog's distribution on the Balkans is scarce and unreliable. The unconfirmed collected records could be validated by extensive camera trap studies. It is vital to include local people (particularly hunters) in monitoring the raccoon dog's invasion. For this purpose, they need to be properly trained to identify the species and the signs of its presence. Currently, most of the hunters in the Balkan countries have difficulty identifying the species. A comprehensive educational campaign is essential for the Balkans, since most of the people are not aware of the raccoon dog's presence in their countries. Along with systematic research, this would be the important first step towards understanding the scope of the invasion and potential effects. Without this proper scientific foundation, no management plans and actions can be undertaken.

REFERENCES

1. Adamopoulou C. and Legakis A., 2016 – First account on the occurrence of selected invasive alien vertebrates in Greece, *BioInvasions Records*, 5, 189-196.
2. Barrat J., Richomme C. and Moinet M., 2010 − The accidental release of exotic species from breeding colonies and zoological collections, *Revue Scientifique et Technique (International Office of Epizootics)*, *29*, 1, 113-22.
3. Catsadorakis G. and Bousbouras D., 2010 − The mammalian fauna: an annotated list. In G. Catsadorakis and H. Kallander (Eds.), The Dadia-Lefkimi-Soufli Forest National Park, Greece: Biodiversity, management and conservation, 207-214). Athens: WWF Greece.
4. Ćirović D., 2006 − First record of the raccoon dog (Nyctereutes procyonoides Gray, 1834) in the former Yugoslav Republic of Macedonia, *European Journal of Wildlife Research*, *52*, 2, 136-137.
5. Cirovic D. and Milenkovic M., 1999 − Previous findings of the raccoon dog (Nyctereutes procyonoides ussuriensis Matschie 1907) in Yugoslavia and analysis of probable paths of its immigration, *Contributions to the Zoogeography and Ecology of the Eastern Mediterranean Region*, 1, 77-82.
6. Dragoev P., 1978 − Obogatyavane na lovnata fauna v Bulgaria [Enrichment of the game fauna in Bulgaria], Zemizdat, 102. (in Bulgarian)
7. Drygala F., Stier N., Zoller H., Boegelsack K., Mix H. M. and Roth M., 2008 − Habitat use of the raccon dog (Nyctereutes procyonoides) in north-eastern Germany, *Mammalian Biology*, 73, 5, 371-378.
8. Duplić A., Slijepčević V., Popović N. and Jedriško P., 2016 – First record of the raccoon dog (Nyctereutes procyonoides) in the Croatian parts of Dinarides. In Book of Abstracts of the 2nd Croatian symposium on invasive species/Jelaska, Sven D. (ur.). - Zagreb: Croatian Ecological Society, 50.
9. ESRI, 2011 − ArcGIS Desktop. Redlands, CA: Environmental Systems Research Institute.
10. Genov P., 2012 − Poznavame li enotovidnoto kuche (Nyctereutes procyonoides Gray.). [Do we know the raccoon dog (Nyctereutes procyonoides Gray)], *Lov I Ribolov*, 10, 74-78. (in Bulgarian)
11. Georgiev D., 2010 − Species composition, numbers and influence of predatory mammals on breeding waterbirds in protected area "Pomorie lake." In Proceedings of the Integrated management plan for protected sites "Pomorie lake" BG0000152 and „Pomorie" BG0000620, 141-144. (in Bulgarian)
12. Green Balkans, 2001 − Biodiversity in the area of Belene Island, Bulgaria, Report on Item 6 of the Terms of Reference under Project Agreement between Green Balkans and WWF Greece, 2000, 27.
13. Kauhala K. and Kowalczyk R., 2011 − Invasion of the raccoon dog Nyctereutes procyonoides in Europe: History of colonization, features behind its success, and threats to native fauna, *Current Zoology*, 57, 5, 584-598.
14. Kauhala K. and Saeki M., 2004 − Raccoon dogs: Finnish and Japanese raccoon dogs – on the road to speciation? In D. W. Macdonald and C. Sillero-Zubiri (Eds.), Biology and Conservation of Wild Canids, 216-226, Oxford, United Kingdom: Oxford University Press.
15. Kauhala K. and Saeki M., 2016 − Nyctereutes procyonoides. The IUCN Red List of Threatened Species 2016: e.T14925A85658776. http://dx.doi.org/10.2305/IUCN.UK.2016-1.RLTS.T14925A85658776.en
16. Milačič R., Ščančar J. and Punović M., (eds), 2015 − The Sava River, The Handbook of Envirnmental Chemistry, Springer-Verlag Berlin Heidelberg. ISBN 978-3-662-44034-6, DOI 10.1007/978-3-662-44034-6-1.
17. Peshev T. and Yordanov M., 1968 − Enotopodobnoto kuche. Nov bozaynik za bulgarskata fauna/The raccoon dog. New mammal for the Bulgarian fauna, *Lov I Ribolov*, 8, 5. (in Bulgarian)

BARBUS MERIDIONALIS RISSO, 1827 POPULATIONS STATUS IN THE VIŞEU RIVER BASIN (MARAMUREŞ MOUNTAINS NATURE PARK)

Doru BĂNĂDUC * and *Angela CURTEAN-BĂNĂDUC* **

* "Lucian Blaga" University of Sibiu, Faculty of Sciences, Natural Sciences Department, Dr. Ion Raţiu Street 5-7, Sibiu, Sibiu County, Romania, RO-550012, ad.banaduc@yahoo.com
** "Lucian Blaga" University of Sibiu, Applied Ecology Research Center, Dr. Ion Raţiu Street 5-7, Sibiu, Sibiu County, Romania, RO-550012, angela.banaduc@ulbsibiu.ro, ad.banaduc@yahoo.com

DOI: 10.1515/trser-2017-0016

KEYWORDS: Human impact, habitats, evaluation, Romania.

ABSTRACT

The ecological state of lotic ecosystems occupied naturally by *Barbus meridionalis,* in the Vişeu Basin within the Maramureş Mountains Natural Park, vary among good to reduced. The inventoried human activities which negatively influence the ecologic state of the *Barbus meridionalis* species habitats and populations are the organic and mining pollution, and poaching. The habitats with low and inadequate conditions created a reduced status of the *Barbus meridionalis* populations; the status of *Barbus meridionalis* populations is not so much affected in the cases of habitats of average to good condition. *Barbus meridionalis* is considered a relatively common fish species in the researched watershed despite the fact that its populations ecological status has decreased from 2007-2015, but the restoration potential in the area for improving this species status is high.

RESUMEN: Estado de las poblaciones de *Barbus meridionalis* Risso 1827, en la cuenca del río Vişeu (Parque Natural Montañas Maramures).

El estado de los ecosistemas lóticos que ocupa de forma natural *Barbus meridionalis* en la cuenca Vişeu, dentro del Parque Natural Montañas Maramures, varía entre bueno a deteriorado. El inventario de actividades humanas que tienen un efecto negativo sobre el estado del hábitat y las poblaciones de *B. meridionalis*, incluyen la contaminación orgánica y por minería y la pesca furtiva. Los hábitats cuyas condiciones son inadecuadas para la especie, reducen el tamaño de las poblaciones de *B. meridionalis.* Las poblaciones que se encuentran en hábitats en regular o buen estado de conservación, no son afectadas significativamente. Esta especie es considerada como un pez común en la cuenca, a pesar de que el estado de sus poblaciones se ha deteriorado de 2007 a 2015; existe, sin embargo, un gran potencial de restauración en el área como para mejorar el estado de la especie.

REZUMAT: Starea populaţiilor de *Barbus meridionalis* Risso, 1827, în bazinul râului Vişeu (Parcul Natural Munţii Maramureşului).

Starea ecologică a ecosistemelor lotice ocupate în mod natural de *Barbus meridionalis* în bazinul Vişeu, din Parcul Natural Munţii Maramureşului, variază între bună şi slabă. Activităţile umane inventariate, care influenţează negativ starea ecologică a habitatelor şi populaţiilor speciei *Barbus meridionalis* sunt poluarea organică şi mineritul şi braconajul. Habitatele cu condiţii ecologice inadecvate induc o stare slabă a populaţiilor de *Barbus meridionalis*; starea populaţiilor de *Barbus meridionalis* nu este atât de afectată, în cazurile habitatelor cu o stare medie spre bună. *Barbus meridionalis* este considerată o specie relativ comună în bazinul studiat, în ciuda faptului că starea ecologică a populaţiilor acesteia a scăzut în perioada 2007-2015, dar potenţialul de restaurare a habitatelor în zonă, pentru îmbunătăţirea stării acestei specii, este ridicat.

INTRODUCTION

Most of the Maramureș Mountains Nature Park's streams and rivers are situated in the Vișeu Basin and there are hardly any in the Bistrița Aurie Basin, in the northern Romanian Carpathians (Chiş, 2008; Costea, 2008).

The Vişeu River is one of the principal tributaries of the Danube River, entering into the much bigger Tisa River. It is over 80 km in length and has a multiannual regular discharge of 30.7 m^3/s at its lower part in the proximity at its confluence with the Tisa. The origin is located in the Prislop Pass (1,416 m) and it flows into the Tisa River, in near the locality Valea Vișeului, the basin cover-up a surface of 1,606 km^2. (Ujvari, 1972)

The rather big diversity of lotic and lenitic ecosystems, and their protected species in the Vişeu Watershed are very important from the conservation perspective. The fish are not excluded from this situation, as noted by a variety of ichthyologists in the last century and more. Over 50% of the fish species existing in the studied Maramureș Mountains Nature Park are of protection importance: *Eudontomyzon danfordi* Regan, 1911, *Thymallus thymallus* (Linnaeus, 1758), *Leuciscus souffia* (Risso, 1827), *Romanogobio uranoscopus* (Agassiz, 1828), ***Barbus meridionalis* Risso, 1827,** *Sabanejewia aurata* (De Filippi, 1863), *Cottus gobio* Linnaeus, 1758, and *Hucho hucho* (Linnaeus, 1758). (Bănărescu, 1964; Staicu et al., 1998)

The distribution of *Barbus meridionalis* (Actinopterygii, Cypriniformes, Cyprinidae, Barbinae) is contained in the Danube, Nistru/Dniester, Odra, Vistula and Vardar watersheds. It is also present in the Romanian hydrographic basins, and also in its neighboring countries, but not only: Hungary, Serbia, Croatia, Bulgaria, Moldavia and Ukraine (Bănăduc, 2011; Bănăduc et al., 2012; Cakic et al., 1998; Lenhardt et al., 1996; Moșu et al., 2006; Velykopolsky and Didenko, 2010; Guti, 1995; Trichkova et al., 2009).

Barbus meridionalis is included in the Habitats Directive (92/43/EC) Annex II. In Eastern and Central Europe, it is a quite common species with a good umbrella species potential; a similar situation exists in the Romanian hydrographic net as well. This species is a lithophilic, reophilic and benthopelagic freshwater fish that lives in ecosystems with water temperatures below 25˚C in streams located over 500 m above sea level. It is a short-living species in mountainous, tableland and a few lowland rivers with appropriate ecosystems. It is favoured by fast flowing and clear aquatic sectors and hard lithologic substrata. In the second or third year of life it reaches reproduction age. Reproduction happens in the spring season; every now and then is extended until the summer season, (from May to July). Along the reproduction period, they congregate in flocks and are in motion upstream and seek good gravel and stones substrata. The food of alevines reside principally of benthic invertebrates (trichopterans, ephemeropterans, tendipedes, gamarids, oligochetes, etc.) and plant litter. The adults feed on these macroinvertebrates too and with fries and alevines (Baensch and Riehl, 1995; Kottelat and Freyhof, 1972; Bănăduc et al., 2011).

The preservation measures for this species should aim for a favorable conservation status, which should mirror a good equilibrium of the total pressures influencing this species that can alter its long-term life quality. In this context, particular actions should be identified and proposed for the specific situation of the study area. In the context in which the human impact is one of the main worldwide determinants that cause structural alterations in fish associations (Bănăduc et al., 2016; Breine, 2011; Halpern et al., 2015), the central aim of this research is to evaluate the conservation status of the *Barbus meridinalis* populations living in the Maramureș Mountains Nature Park (Eastern Carpathians) and the particular results are to bring to light the site management elements for improving the ecological condition of these populations.

MATERIAL AND METHODS

Research on the *Barbus meridionalis* populations of the Maramureş Mountains Natural Park were done in 2007-2015, and included 370 sampling lotic sectors (Fig. 1). This research consisted of studied species mapping, evaluation of the present preservation status, and characterization of the elements which induce the actual populations status.

The research is based on the working hypothesis (a) and null hypothesis (b): 1a. Aquatic habitats with low ecologic conditions that have reduced the *Barbus meridionalis* populations; 1b. There will be no variation in the *Barbus meridionalis* populations between habitats of reduced, average or good condition; 2a. The populations of *Barbus meridionalis* has decreased along the period 2007-2015; the population of *Barbus meridionalis* has not decreased in the same period.

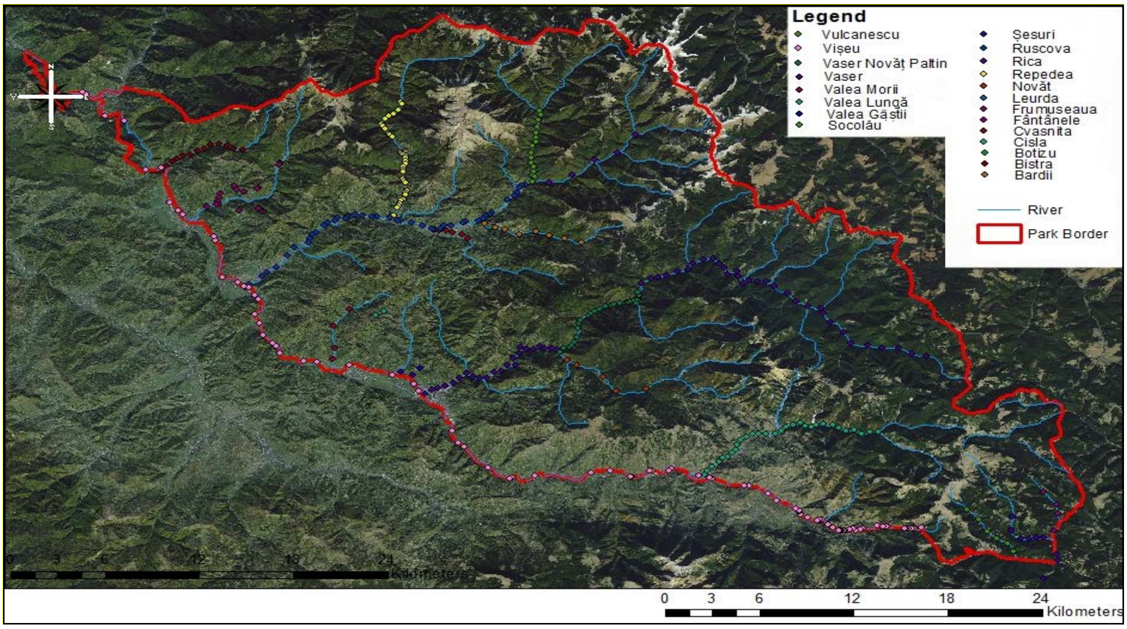


Figure 1: The 370 sampling stations location; GIS support Danci O.

To evaluate the *Barbus meridinalis* populations' status in the Maramureş Mountains Nature Park, quantitative samples were taken from sampling stations within a three kilometers range between two successive stations on all potential habitats with suitable environments for the studied fish species. The locations of the sampling stations admit the evaluation of the effects of the human impact on the researched fish populations, containing the biotope characteristics changing, riverbed exploitation, hydrotechnical works, pollution sources, unrestrained sport fishing and poaching.

Fish fauna quantitative sampling was done through electronarcosis, per time and effort unit, and per each researched lotic section (two hours on the Vişeu River, one hour on Ruscova, and 30 minutes on the other smaller rivers), on five longitudinal sectors of 100 m length. After the fish identification, all fish were immediately released back in their habitat.

The number of fish sampled in the time/effort unit in each station can be converted by correlation in the following fish species classes: (C) – common, (R) – rare, or (V) – very rare, like in the Natura 2000 standard data form filling guidelines, "In mammals, amphibians, reptiles and fishes, no numeric information can be indicative and then the size/density of the population is evaluated as common species – (C), rare – (R), or very rare species – (V)".

The elements used to evaluate the studied fish population statuses are: population size, areal size, balanced sharing of individuals by classes of age, and the proportion of fish individuals of *Barbus meridionalis* in the local fish associations structure.

Analogous to the Natura 2000 guidance, standard data from the criteria "The conservation degree of specific habitats", and the subcriteria: i) the degree of conservation of the habitat features which are important for the species; ii) possibilities for recovery.

The criterion i) requires a total assessment of the typical features of the habitat concerning the needs of the studied species. "The best expertise", is applied to rank these criteria: I. elements in excellent condition, II. well preserved elements, III. elements in moderately or partially degraded condition.

In the situation in which the subclass I is granted "I: elements in excellent condition" or "II: well preserved elements", the criteria B (b) should be grouped as "A: excellent conservation", or "B: good conservation", indifferent of the other sub-criteria.

In the case of the sub-criterion ii) which is considered exclusively if the items are partially deteriorated, an evaluation of the studied population viability is necesary. The achieved classification system is: I. not difficult recovery; II. possible restoration with average effort; III. questionable or unattainable restoration.

The procedure for classification is based on the sub-criteria: A – excellent conservation = elements in excellent condition, indifferent of classification of the possibility for recovery; B – good conservation = elements in moderate or incompletely degraded condition and not difficult to restore; C – moderate or decreased conservation = all other mixtures.

In all researched areas, the following was evaluated: condition, pressures/threats of habitats and populations of *Barbus meridionalis*.

The studied lotic sections to evaluate the conservation status of *Barbus meridionalis* in the researched basin were approached in areas where the studied populations are permanent, with the potential of well preserved typical habitats, as well as lotic sectors located at the borderline of the local studied basin range area for the studied fish species, which include sectors where human activities can present danger in the local populations' status – the Representativity Criteria.

Barbus meridionalis Riso, 1827 (Natura 2000 Code 1138), (RO − moioagă, moiţă, cîrcuşă, jumugă, jamlă, jamnă, mreană pătată, mreană vânătă, mreană de munte, mreană de vale; BG − Cherna, DE − Forellenbarbe, Semling, Afterbarbe; FR − Barbeau truite, Truitat, Turquan; GB − Mediterranean barbell; HU − Petenyi-márna; CS − Potocna mrena) with its terra typica in the Mureş River in Transylvania/Romania has the following general descriptive elements: elongated body; the superior body profile is an ascendant curveline from the snout to the dorsal fin, without reaching the dorsal fin; the last simple radia of the dorsal fin is thin, flexible and not jagged; the ventral fins are inserted backward to the dorsal fin insertion; the dorsal fin edge is plain or slightly fluted; the lips are relatively fleshy and developed; the posterior whiskers are sometimes long, exceeding the eye; the back of the body is dark brown-rusty colored, with darker and lighter spots, the flanks are yellow-rusty with spots, the ventral side is light yellow; the dorsal and caudal fins have accentuated spots, the rest of the fins are yellowish; the whiskers are yellowish with no red axis; and it can reach 28-30 cm in length. The general ecologic elements are: benthopelagic and freshwater fish; a short-lived species which is found in mountainous and hilly rivers, with springs in this area; prefer the clear and fast flowing water sectors and the hard substrata; no migrations were registered; the reproduction happens in the spring, sometimes is prolonged until the summer; its food consists mainly of benthic aquatic invertebrates (tendipedes, ephemeropterans, trichopterans, gamarids, oligochetes and rarely plants) (Bănărescu, 1964; Bănărescu and Bănăduc, 2007).

RESULTS

The river sectors where *Barbus meridionalis* (Fig. 2) was sampled during the study period are presented in table 1 (Fig. 4), for the studied lotic sectors, the catch index values were offered in the paper (individual numbers per time and effort unit.)

Figure 3: Sampled *Barbus meridionalis* Risso, 1827 individuals.

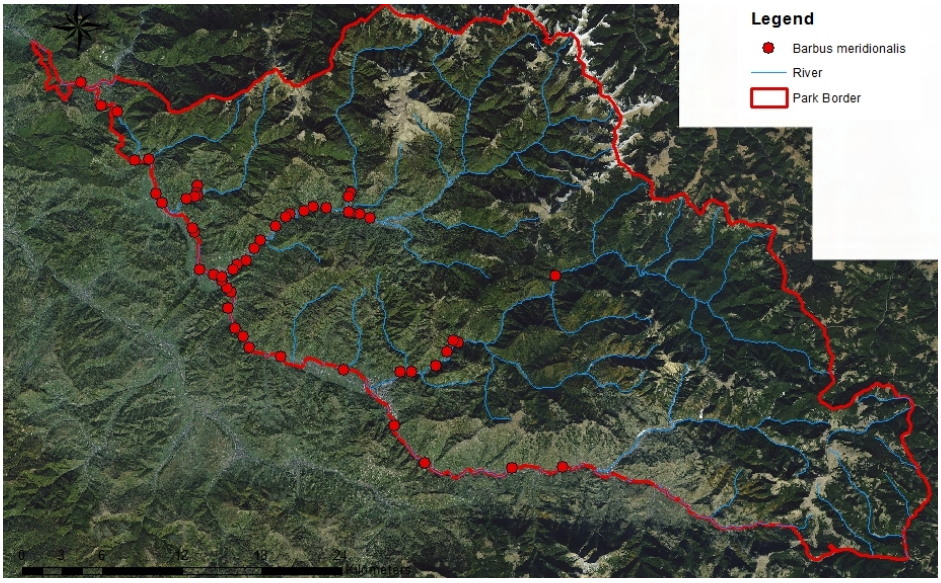


Figure 4: Sampling stations location where *Barbus meridionalis* was found in Vişeu Basin/ Maramureş Mountains Nature Park; GIS support Danci O.

Table 1: *Barbus meridionalis* sampling points in the study area.

No. crt.	Lotic system	Station code	Lat. (N’)	Long. (E’)	Catch index no. ind./100 m × 30 min	Characteristic habitat state
1.	Vișeu River	39	47 39 30.9	24 37 03.9	1	reduced
2.	Vișeu River	43	47 39 26.1	24 34 0.,3	1	reduced
3.	Vișeu River	47	47 39 36.6	24 28 49.4	1	reduced
4.	Vișeu River	50	47 41 07.4	24 26 57.0	1	reduced
5.	Vișeu River	53	47 43 24.0	24 23 53.7	1	reduced
6.	Vișeu River	55	47 43 54.9	24 20 04.5	3	reduced
7.	Vișeu River	57	47 44 16.2	24 18 11.5	1	reduced
8.	Vișeu River	59	47 44 41.5	24 17 49.2	6	reduced
9.	Vișeu River	60	47 45 01.4	24 17 20.4	12	average
10.	Vișeu River	62	47 45 52.5	24 16 53.8	2	average
11.	Vișeu River	63	47 46 29.7	24 17 03.6	32	average
12.	Vișeu River	64	47 46 40.3	24 16 52.0	42	good
13.	Vișeu River	65	47 46 58.9	24 16 32.4	39	good
14.	Vișeu River	67	47 47 11.3	24 16 01.2	26	good

Table 1 (continued): *Barbus meridionalis* sampling points in the study area.

No. crt.	Lotic system	Station code	Lat. (N')	Long. (E')	Catch index no. ind./100 m × 30 min	Characteristic habitat state
15.	Vișeu River	68	47 47 23.3	24 15 11.8	11	good
16.	Vișeu River	69	47 48 55.0	24 14 50.1	9	good
17.	Vișeu River	70	47 49 04.9	24 14 44.8	7	good
18.	Vișeu River	72	47 50 07.4	24 12 54.1	36	good
19.	Vișeu River	73	47 50 27.7	24 12 29.2	45	good
20.	Vișeu River	74	47 51 53.1	24 12 03.0	52	good
21.	Vișeu River	75	47 51 49.4	24 11 12.2	55	good
22.	Vișeu River	76	47 53 47.5	24 10 06.7	56	good
23.	Vișeu River	77	47 54 00.3	24 09 07.7	48	good
24.	Vișeu River	79	47 54 58.5	24 07 56.4	50	good
25.	Repedea River	29	47 50 36.1	24 24 14.8	5	good
26.	Repedea River	31	47 50 24.8	24 24 07.9	7	good
27.	Vaser River	37	47 44 30.6	24 30 44.0	3	average
28.	Vaser River	38	47 44 36.0	24 30 26.2	9	average

Table 1 (continued): *Barbus meridionalis* sampling points in the study area.

No. crt.	Lotic system	Station code	Lat. (N’)	Long. (E’)	Catch index no. ind./100 m × 30 min	Characteristic habitat state
29.	Vaser River	40	47 44 09.1	24 30 04.0	6	average
30.	Vaser River	43	47 43 35.3	24 29 26.7	12	average
31.	Vaser River	47	47 43 19.5	24 27 58.3	21	average
32.	Vaser River	49	47 43 19.3	24 27 18.1	8	average
33.	Novăţ Stream	31	47 47 15.0	24 36 35.6	2	average
34.	Ruscova River	19	47 49 34.5	24 25 24.3	1	reduced
35.	Ruscova River	20	47 49 42.7	24 24 47.8	1	average
36.	Ruscova River	21	47 49 46.8	24 24 16.3	1	average
37.	Ruscova River	22	47 49 47.7	24 24 05.6	3	average
38.	Ruscova River	25	47 49 56.6	24 22 46.3	7	average
39.	Ruscova River	27	47 49 59.4	24 21 58.5	5	average
40.	Ruscova River	30	47 49 49.7	24 21 25.4	11	average
41.	Ruscova River	33	47 49 42.5	24 20 33.9	13	average
42.	Ruscova River	34	47 49 33.8	24 20 20.9	9	average
43.	Ruscova River	36	47 49 1.,0	24 19 45.5	4	average
44.	Ruscova River	39	47 48 36.9	24 18 50.8	13	average
45.	Ruscova River	40	47 48 18.0	24 18 29.4	15	average

Table 1 (continued): *Barbus meridionalis* sampling points in the study area.

No. crt.	Lotic system	Station code	Lat. (N')	Long. (E')	Catch index no. ind./100 m × 30 min	Characteristic habitat state
46.	Ruscova River	41	47 47 47.5	24 17 58.4	6	average
47.	Ruscova River	42	47 47 36.6	24 17 27.1	8	average
48.	Ruscova River	43	47 47 25.6	24 17 11.0	22	average
49.	Ruscova River	44	47 47 07.0	24 16 32.0	15	average
50.	Frumuşeaua River	12	47 50 49.1	24 15 00.5	1	average
51	Frumuşeaua River	14	47 50 31.4	24 14 55.8	1	average
52.	Frumuşeaua River	16	47 50 24.3	24 14 58.8	4	average
53	Frumuşeaua River	17	47 50 23.0	24 14 51.3	3	reduced
54.	Frumuşeaua River	18	47 50 16.7	24 14 21.4	2	reduced

DISCUSSION

Based on the results of this research, and consistent with the *Barbus meridionalis* fish species' ecological and biological necessities and local status, three risk elements (pressures and threats): organic pollution, poaching and pollution resulted from mining activities were found.

Organic pollution coming from sewage systems, agriculture and fish farms have a negative impact on the Vişeu River basin ichthyofauna. It is a persistent and bad situation connected to improper sewage systems and inefficient wastewater treatment; also to farms, in the Vișeu Basin, mainly on the Vișeu River, situation which are a permanent negative impact on fish fauna (Oprean et al., 2009).

Poaching. During the field research, around the clock poaching activities (in over 20% of our field trips) using a great variety of home-made electrofishing gears were observed. Also, poachers were observed during their illegal activities using a large variety of substances for killing and collecting fish of all dimensions. By asking 431 local people in the Maramureș Mountains Nature Park, it seems that poaching is a frequent activity in all seasons in the Vișeu Basin, and this induces an important diminishing of the *Barbus meridionalis* abundance and modifications of the age classess structure of the local populations.

Mining activities pollution. The long term pollution resulting from heavy metal mining and storage activities in an old mining industry area (Fig. 7) of Ţâşla Stream basin are negatively influencing not only the Ţâşla lotic aquatic habitats, but also the habitats and species of interest of the upstream Vișeu River; the effects of the precipitates washing from waters of the mine galleries and greened refuse heaps is a heavy one in the Ţâşla Stream basin and is also serious on the upstream Vişeu River (Staicu et al., 1998). The synergism among the identified human impact puts pressure on numerous lotic sectors in the researched area (Figs. 5 and 6) and the evaluation score for the researched fish species is not at the natural potential.

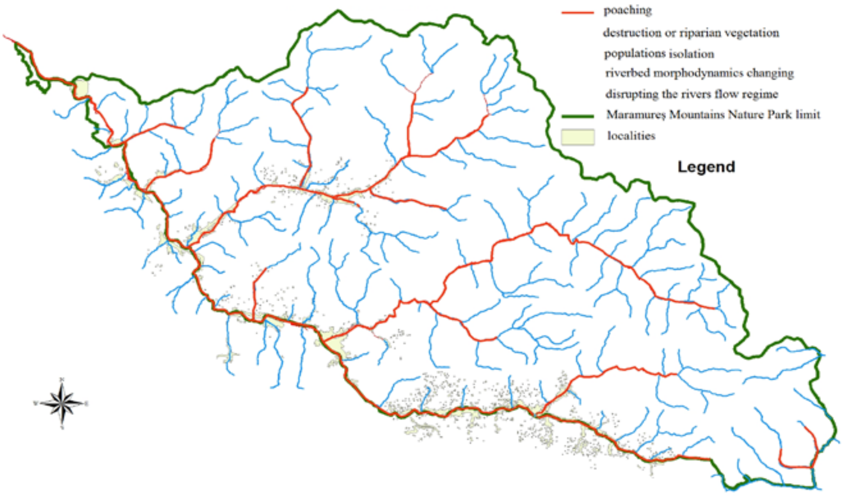


Figure 5: Diagnosed combined pressures and threats for *Barbus meridionalis* in the studied Vişeu River basin/Maramureş Mountains Nature Park area.

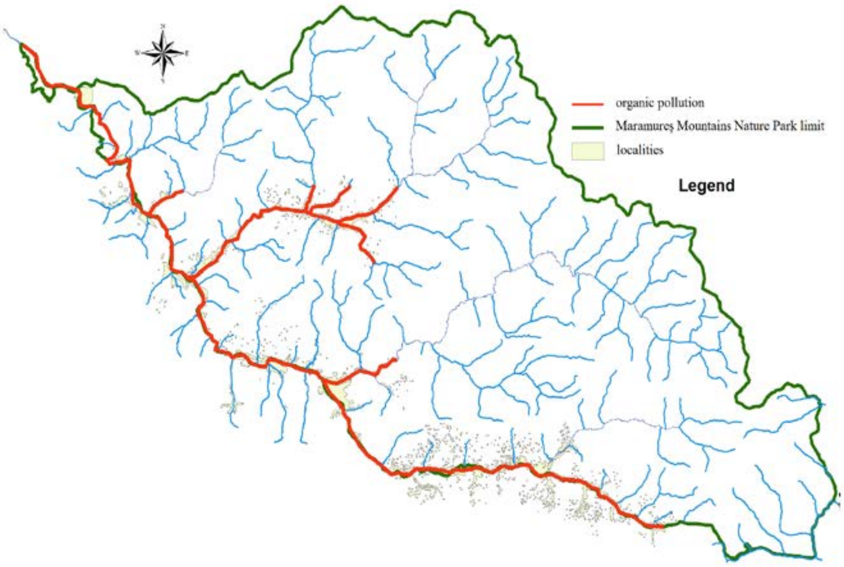


Figure 6: Lotic sectors influenced by organic pollution; in the studied Vişeu River basin/Maramureş Mountains Nature Park area.

Figure 7: Dissafected mining industrial area and refuse heaps in the Tâşla Stream proximity.

There are some main management measures necessary for the diminishing or elimination of the negative effects of the identified pressures and threats for the *Barbus meridionalis* populations in Vişeu Basin in Maramureş Mountains Nature Park.

Organic pollution. Sewerage systems must be developed throughout all of the Vişeu Basin and also the domestic and zootechnical waters should be properly cleaned in all the basin localities.

Poaching. It is proposed to increase the number of hours of on-site verification of potential poaching activities by the local protected area rangers. The lack of financial resources for these activities can be compensated, at least in part, through permanent cooperation agreements with forestry, police, and gendarmerie, etc. personel, as well as by creating a permanent structure of local volunteers.

Mining activities pollution. The impact of mine drainage and tailing dumps washing can be significantly reduced by sealing existing mine galleries and renaturation/isolation of mine tailing dumps in the Ţâşla River basin.

Finally, it is apparent that the aquatic habitats' quality influenced the *Barbus meridionalis* populations: the habitats with low ecologic conditions have reduced the status of *Barbus meridionalis* populations; the status of *Barbus meridionalis* populations is not so much affected in the cases of habitats of average to good conditions. In general, the populations' status of the *Barbus meridionalis* has decreased along the period of 2007-2015.

CONCLUSIONS

Barbus meridionalis Risso, 1827 is one of the fish species of valuable preservation responsibility within the Vişeu Basin, one of the isolated areas in Northern Romania. The status of aquatic ecosystems naturally inhabited by *Barbus meridionalis,* within the Maramureş Mountains Nature Park, oscillates in the best cases among good (27.78% of the lotic sampled sections with *Barbus meridionalis*), average (51.85%), and reduced (20.37%.) Excellent conservation status is now missing for populations of this species in the studied basin.

The determined human impact types (organic pollution, poaching and mining activities pollution), diminish the *Barbus meridionalis* habitat's ecologic state and as a result the studied fish species populations, are under their natural potential.

Barbus meridionalis has permanent populations in the studied area, but in their natural potential, in comparison with historical data due to last half of a century, human impact is not fulfilled in terms of aquatic habitat quality and abundance of the studied fish species individuals; in the Vișeu in the upper and middle sectors, the statuses vary from upstream to downstream from reduced to average, and in the middle and lower sectors, the statuses vary from average to good. The sampling sections of the Repedea River fall under the good status, Vaser River is under the average status, Novăţ Stream is also under the average status, the Ruscova River is under the average status, and the Frumuşeaua River is under the average-reduced status. The habitats of this fish species are large enough as total surface, with relatively good lotic connections within the studied basin to preserve the present ecological state of the Mediterannean barbell species populations.

Based on this research, *Barbus meridionalis* is a relatively common fish species in the Vişeu River basin, and there, where it is at its natural potential in the present, a restoration potential is not difficult to be reached (upper-middle Vișeu River, Vaser River, Novăţ Stream, Ruscova River, and Frumuşeaua River).

ACKNOWLEDGEMENTS

These data were obtained in the project "Inventarierea, cartarea și evaluarea stării de conservare a speciilor de pești din Parcul Natural Munții Maramureșului (ROSCI 0124 Munții Maramureșului)/Inventory, mapping and assessment of the conservation status of fish species of Munții Maramureșului Nature Park (ROSCI 0124 Maramureșului Mountains)". Thanks for the GIS support to Ms. Danci O. Special thanks for the continuous support of the Munții Maramureșului Natural Park Administration and Scientific Council members especially to: Bogdan C., Bucur C., Szabo B., Brener A. and Mărginean M.

REFERENCES

1. Baensch H. A. and Riehl R., 1995 – Aquarien Atlas, Band 4, Mergus Verlag GmbH, Verlag für Natur-und Heimtierkunde, Melle, Germany, 864.
2. Bănăduc D., 2011 – New SCIS proposal regarding the ichtiofauna after the Alpine Biogeographic Seminar for Romania, Sibiu (Transylvania, Romania) 9-12 June 2008, *Acta Oecologica Carpatica*, IV, 175-184.
3. Bănăduc D. and Curtean-Bănăduc A., 2013 – Barbus meridionalis Risso 1827 (syn. Barbus balcanicus) monitoring elements proposal for Croatia, in Natura 2000 context, *Transylvanian Review of Systematical and Ecological Research*, 15.1, The Wetlands Diversity, 163-182.
4. Bănăduc D., Nagy A. and Curtean-Bănăduc A., 2012 – New SCIS proposal regarding the ichtiofauna after the Continental Biogeographic Seminar for Romania, Sibiu (Transylvania, Romania) 9-12 June 2008, Acta Oecologica Carpatica, V, 145-162.
5. Bănăduc D., Oprean L., Bogdan A. and Curtean-Bănăduc A., 2011 – The analyse of the trophic resources exploitation by the congeneric species Barbus barbus (Linnaeus, 1758) and Barbus meridionalis Risso 1827 in the Târnava River basin (Transylvania, Romania), *Transylvanian Review of Systematical and Ecological Research*, 12, The Wetlands Diversity, 101-110.
6. Bănăduc D., Rey S., Trichkova T., Lenhardt M. and Curtean-Bănăduc A., 2016 – The Lower Danube River-Danube River-Danube Delta-North West Black Sea: A pivotal area of major interest for the past, present and future of its fish fauna – A short review, *Science of the Total Environment*, 545-546, 137-151.
7. Breine J., Stevens M., Van den Bergh E. and Maes J., 2011 – A reference list of fish species for a heavily modified transitional water: Zeeschelde, *Belgian Journal of Zoology*, 141, 1, 44-55.
8. Cakic D., Hegediš A., Kataranovski D. and Lenhardt M., 1998 – Endohelminths of Mediterranean barbel, Barbus peloponnesius petenyi, in running waters of West Serbia (Yugoslavia), *Folia zoologica*, 47 (Suppl 1), 81-85.
9. Chiș T. V., 2008 – Gegraphical background elements of the Vișeu River basin (Maramureș, Romania), *Transylvanian Review of Systematical and Ecological Research*, 5, The Maramureș Mountains Nature Park, 1-4.
10. Costea M., 2008 – The Characteristic of the hydrographical basins of the Maramureș Mountains (Maramureș, Romania), *Transylvanian Review of Systematical and Ecological Research*, 5, The Maramureș Mountains Nature Park, 13-20.
11. Guti G., 1995 – Conservation status of fishes in Hungary, *Opusc Zool Budapest*, XXVII-XXVIII, 153-158.
12. Halpern B. S., Frazier M., Potapenko J., Casey K. S., Koening K., Longo C., Lowndes S., Rockwood R. C., Saeling E. R., Selkoe K. A. and Walbridges S., 2015 – Spatial and temporal changes in cumulative human impacts on the world's ocean, *Nature Communications*, 6.
13. Kottelat M. and Freyhof J., 1972 – Handbook of European freshwater fishes, Publications Kottelat, Cornol and Freyhof, Berlin, 646.
14. Lenhardt M., Mickovic B. and Jakovcev D., 1996 − Age, growth, sexual maturity and diet of the Mediterranean barbel (Barbus peloponnesius petenyi) in the river Gradac (West Serbia, Yugoslavia), *Folia Zoologica*, 45, (Suppl 1), 33-37.

15. Moșu J. A., Davideanu G. G. and Cebanu A., 2006 – Elements of the Ichthyofauna diversity of Prut River basin, *Acta Ichtiologica Romanica*, I, 171-184.
Oprean L., Curtean-Bănăduc A. and Bănăduc D., 2009 – Vişeu river watershed (Maramureş, Romania) ecological management proposal, Management of Sustainable Development, 1, 31-39.
16. Staicu G., Bănăduc D. and Găldean N., 1998 − The structure of some benthic macroinvertebrates and fishes communities in the Vişeu Watershed, Maramureş, Romania, *Travaux du Museum National d'Histoire Naturelle "Grigore Antipa"*, Bucureşti, 587-608, vol. XL.
17. Trichkova T., Stefanov T., Vassilev M. and Zivkov M., 2009 – Fish species diversity in the rivers of north-west Bulgaria, *Transylvanian Review of Systematical and Ecological Research*, 8, The Wetlands Diversity, 161-168.
18. Ujvari I., 1972 – Geografia apelor României, Ed. Ştiinţifică, Bucureşti, 590. (in Romanian)
19. Baensch H. A. and Riehl R., 1995 – Aquarien Atlas, Band 4, Mergus Verlag GmbH, Verlag für Natur-und Heimtierkunde, Melle, Germany, 864.
20. Velykopolsky I. I. and Didenko A. V., 2010 – Suchasnyi stan zapasiv mareny karpats'koi u richkah Zakarpattia z ogliadu ii pryrodookhoronnogo statusu, Fisheries Science of Ukraine, 4, 14, 51-58. (in Ukrainian)