ANALELE ȘTIINȚIFICE
ALE
UNIVERSITĂȚII „ALEXANDRU IOAN CUZA”
DIN IAȘI
(SERIE NOUĂ)

S E C Ț I U N E A I

BIOLOGIE ANIMALĂ

Supliment

PROCEEDINGS OF THE INTERNATIONAL SCIENTIFIC CONFERENCE

THE ANTHROPOGENIC IMPACT UPON BLACK SEA REGION BIODIVERSITY

dedicated to celebrating 85 years since the establishment of „Prof. dr. Ioan Borcea” Marine Biology Research Unit – Agigea, „Alexandru Ioan Cuza” University of Iaşi

Agigea – Constanta, the 21st-22nd of October 2011

Editura Universităţii „Alexandru Ioan Cuza” Iaşi
2013

Editorial address:

Facultatea de Biologie
Universitatea „Alexandru Ioan Cuza" din Iași
Bd. Carol I, Nr. 20A, 700505 Iași, România

Telephone: +40232201527
Fax: +40232201472

http://www.bio.uaic.ro/publicatii/anale_zoologie/anale_zoo_index.html

SUMMARY

THE STRUCTURE AND LIFEHISTORY OF A POPULATION OF *IDOTEA BALTHICA* FROM AGIGEA AREA

IONELA RALUCA VACAREANU[1] and VICTOR SURUGIU[2]
[1] University of Bucharest, e-mail: v_ralook@yahoo.com
[2] "Alexandru Ioan Cuza" University of Iasi, Faculty of Biology, Bulevardul Carol I, Nr. 20A, Iasi, Romania, e-mail: vsurugiu@uaic.ro

Abstract. In this paper we present the results of a research that focused on lifehistory and structure of the isopod *Idotea balthica* from the Agigea area. The other two species of isopods cited for Romanian Black Sea coast, *Idotea metallica* and *Synisoma capito* were not found among individuals collected. The results indicate that the highest value of biomass was recorded in May (20.4936 $g.m^{-2}$); in the coming months, biomass decreased because of male mortality and recruitment of juvenile in this period. August is characterized by the lowest biomass (1.9467 $g.m^{-2}$) and density 341 $ind.m^{-2}$. The histograms show changes in population size classes for the six months study. Notice the difference between May, where individuals are large, sexually mature and June, July, August, September and October characterized by small to medium sized individuals. The analysis of co-variation of biomass and density values shows that these individuals have two breeding periods represented by the May-June and October. Pregnant females were found in July, August and September, but in a very small number compared with the breeding periods. The number of eggs, embryos or youngs found is also much lower in July-September period. Of 126 pregnant females just one presented two larval stages, egg and embryo, all other larval stage found one, most showing eggs or youngs. *Idotea* is an isomorphic species which can be seen from the analysis of the correlation index.

Key words: *Idotea balthica,* Black Sea

Abstract. Structura şi dinamica unei populaţii de *Idotea balthica* din zona Agigea. În lucrare sunt prezentate rezultatele cercetărilor care au vizat studiul dinamicii isopodului *Idotea balthica* în zona Agigea-Marea Neagră. Celelalte două specii de isopode citate pentru litoralul românesc al Mării Negre, *Idotea metallica* şi *Synisoma capito* nu au fost regăsite printre indivizii colectaţi. Rezultatele obţinute indică că biomasa maximă a fost înregistrată în luna mai (20,4936 $g.m^{-2}$), după care aceasta scade mult în lunile următoare datorită mortalităţii masculilor şi recrutării în această perioadă a indivizilor juvenili. Luna august este caracterizată de o biomasă minimă (1,9467 $g.m^{-2}$), având, totodată, şi densitatea cea mai mică – 341 $ind.m^{-2}$. Histogramele claselor de mărimi evidenţiază evoluţia populaţiei pe perioada celor şase luni de studiu. Se observă diferenţa dintre luna mai, unde indivizii sunt de dimensiuni mari, maturi din punct de vedere sexual şi lunile iunie, iulie, august, septembrie şi octombrie caracterizate de indivizi de talie mică spre medie. Din analiza concomitentă a variaţiei valorilor biomasei şi a densităţii indivizilor reiese ca acestea au două perioade de reproducere reprezentate de lunile mai-iunie şi octombrie. Femele gestante au fost găsite şi în lunile iulie, august şi septembrie, dar într-un număr foarte mic comparativ cu perioadele de reproducere. Numărul de ouă, embrioni sau pui găsiţi este, deasemeni, mult mai mic în intervalul iulie-septembrie. Din 126 de femele gestante doar una prezenta două stadii larvare, de ou si embrion, la toate celelalte am găsit un singur stadiu larvar, majoritatea prezentând ouă sau pui. *Idotea* este o specie izomorfă fapt care se poate observa şi din analiza indicelui de corelaţie.

Cuvinte cheie: *Idotea balthica,* Marea Neagră

Introduction

Idotea balthica is the most common isopod from the Black Sea. It is a benthic species, but accidentally can also be found in plankton, a few miles from the shore, carried by marine currents with marine plants that cling. Abundance is typically low outside these limits (Morris and Mogelberg, 1973; Gutow, 2006)

which indicates a low floating charge over long distances. *Idotea balthica* is almost absent on abiotic surfaces by food shortages (Gutow & Franke 2003; Gutow, 2006). *Idotea balthica* is abundant on floating macroalgae in the German Bight, especially on brown algae of the order Fucales, with average numbers of about 90 adult animals per kg (Franke *et al.*, 1999; Gutow, 2006).

Idotea balthica is a cosmopolitan marine isopod, met in different oceans, seas and lakes, all over the world. It is a widespread species, not only in terms of different degrees of latitude and longitude, but also in terms of salinity variations. It is one of the eurihaline species, it can support and accommodate different concentrations of salinity, from 3.06‰ (freshwater), through brackish water concentration (11.7‰) and came to live in a average of 85‰ salt, which is twice the concentration of Mediterranean waters, where it finds optimal conditions for life (Carausu, 1955).

I. baltica is omnivorous feeding on benthic microalgae, filamentous algae, macroalgae, detritus, small invertebrates and even its conspecifics (Naylor, 1955; Ravanko, 1969; Sywula, 1964; Nicotry, 1980; Robertson & Mann, 1980; Franke & Janke, 1998). Despite its omnivory, *I. baltica* is rather selective within food categories (Salemaa, 1978; Schaffelke *et al.*, 1995). Its dietary choice involves the selection between different algal species (Schaffelke *et al*., 1995; Schramm *et al*., 1996) but likely between different parts of the algal thallus (Salemaa, 1987). In the northern Baltic Sea, however, *F. vesiculosus* was considered as the main source of food for *I. baltica* (Salemaa, 1987). Feeding preferences among the natural algae decrease as follows: *F. vesiculosus* > *Dictyosiphon foeniculaceus* > *Elachista fucicola* > *Pilayella littoralis* > *Enteromorpha intestinalis* > *Ceramium tenuicorne*. The chemical quality of algae is not the major determinant of feeding preferences.

Relationships between predator and prey are also evident in aquatic ecosystems: in freshwater habitats, isopods respond to the presence of predators by burying deeper into the sediment (Huang & Sih, 1991), and in marine habitats, predators (e.g. fish, shrimps and crabs) may increase the use of vegetation cover by mobile invertebrates (Stoner, 1980; Main, 1987) and infuence feeding mode (Skilleter & Peterson, 1994), vertical distribution and migration (Roberts *et al.*, 1989; Kamermans & Huitema, 1994) and survival (Irlandi *et al.*, 1995) of infaunal bivalves (Bostrom & Mattila, 1999).

The growth rate of females is weaker than the male, a faster growth rate in males is explained by the fact that they more frequently shed than females and they grow each shedding, on average, more than females (Carausu, 1955). In the Baltic Sea, females of *Idotea balthica* normally breed only once during their lifespan. Maturation takes place as the oostegites forming the brood pouch develop in May and early June (Haahtela, 1978; Strong & Daborn, 1979; Tuomi *et al*., 1988). The breeding period starts in May when the females are fertilized during the parturial molt. The female lays eggs into its ventral marsupium (brood pouch) and the juveniles are released before the next molt after an incubation period of about 40 days. Juveniles' diet in our study area consists mainly of *C. glomerata* till they reach the physiological size enabling consumption of *F. vesiculosus*, which constitutes the main proportion of the diet of *I. balthica* adults (Salemaa 1979; Tuomi et a., 1988; Jormalainen *et al.*, 2001; Hemmi & Jormalainen, 2004).

Benthic invertebrates are a very important group of organisms in aquatic ecosystems. They are the main processers of organic matter deposited in sediments, thus having an important role in reintroduction to trophic chains of material sedimented, unless these detritus accumulate in the sediment and need a long time to be mineralized and recycled in the trophic chains. *Idotea* is very important in decomposing algae *P. littoralis* and *Cladophora glomerata*. Benthic invertebrates significantly affect the algal decomposition. *Idotea balthica* contribute most to the decomposition of *C. glomerata.* It is believed that grazers act as stabilizers of benthic communities, as filter feeders increase the stress caused by drifting algae (Paalme et al. 2002). In general, grazing is high in summer, mild in autumn and low in spring (Kotta & Kotta, 2002).

In the Black Sea was found in diet of several fish species: *Mullus barbatus ponticus, Corvina umbra, Symphodus (Crenilabrus) tinca, Symphodus (Cr.) quinquemaculatus, Scorpaena porcus, Ophidion barbatum* (Vinogradova & Hirina, in Carausu, 1955).

Material and methods

In order to study the lifehistory and structure of the isopod *Idotea balthica*, specimens were sampled during May-October 2009. Collection was made at intervals of about 30 days, from a single station, set at 200 meters of Agigea fisheries, on natural rocky . Depth at the point of sampling varies between 0 and 50 cm, due to water and rocky on which individuals were taken. Individuals were found mainly on the underside of stones, among algae and other species isopods (*Sphaeroma serratum* and *Sphaeroma pulchellum*), anemones, mussels, amphipodes. The stones were placed in white plastic buckets and then washed thoroughly with seawater to train all individuals in the remaining water in the bucket. Subsequently, individual collection was performed with tweezers and placed in plastic containers, labeled. Conservation material was made at collecting, using 8% formaldehyde solution in seawater. Identification of larval stages to sex and pregnant counting was done under binocular magnifying glass in the laboratory of Hydrobiology. To determine the length of the body, individuals were measured from cephalic extremity to the end of the telson using a binocular micrometer.

Specimens studied were enclosed in individual containers to determine the wet biomass of each individual, was used for conservation technical alcohol. For determination we used the work of Cărăuşu (1955).

Density and biomass have been reported per square meters headband. Biomass was expressed as wet weight. Bodies have been weighing on an analytical electron balance accurate to 0.0001g. *Abundance* (ind.m^{-2}) was calculated by dividing the number of individuals found on the surface studied in the unit – m^2. *Biomass* (g.m^{-2}) is the total weight of individuals present per unit area. Data were processed by statistical methods using the program M.S. EXCEL.

Results and discussion

In the 6 samples monthly collected were found and analyzed 669 individuals of *Idotea balthica*, of whom 398 were females, of which 31.66% were pregnant. The maximum number of pregnant females was found in May (67.60% of

total females), with an average of 81.46 larval stages.ind^{-1}. A month later, the percentage decreased to 59.76% pregnant females with 66.43 larval stages.ind^{-1}. July and August reduced the percentage of pregnant females more than the number of eggs and embryos (the lowest value recorded was 4.17% pregnant in July and 6.15% in August, with 15.75 larval stages.ind^{-1} in July respectively 18.50 larval stages.ind^{-1}. in August), but increased to 26.42% pregnant in September and 22.58% in October, wearing an average number of 13.21 larval stages.ind^{-1} in September, respectively 14.14 larval stages.ind^{-1} in October.

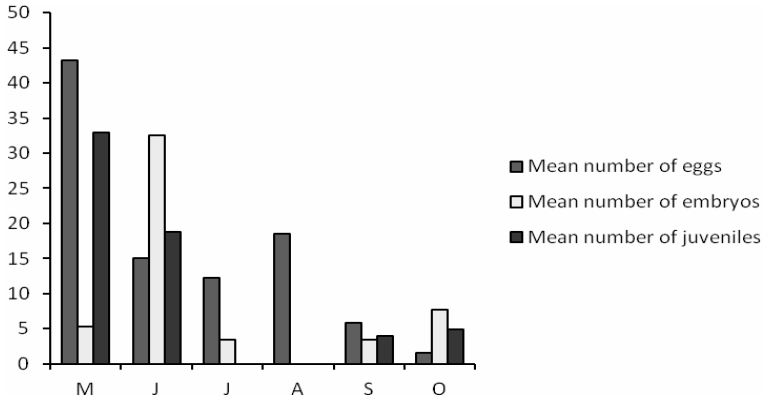


Figure 1. Temporal variation of the average number of eggs, embryos and juveniles in females of *Idotea balthica* from Agigea.

The analysis of the chart (Fig. 1) shows that youngs exceed during breeding periods, the maximum number of young count for a single female is 132 in June and 117 in May.

Maximum monthly average of eggs was 43, value reached in May. A month later this value decreased to 15 eggs per female, reaching minimum values in October (2 eggs per female). In July and August we didn't find youngs.

Of 126 pregnant females only one had two larval stages (egg and embryo), all other females had only one larval stage, most showing eggs or young.

Maximum density was recorded in July 751 ind.m^{-2} (Fig. 2), the density decreased over the coming months and reached 484 ind.m^{-2} in September and 443 ind.m^{-2} in October. The minimum value of 341 ind.m^{-2} for August was due to weather conditions during the sampling of individuals. The sea was rough, the waves were very strong, with a frequency of 3 to 5 seconds and up to 60-80 cm high.

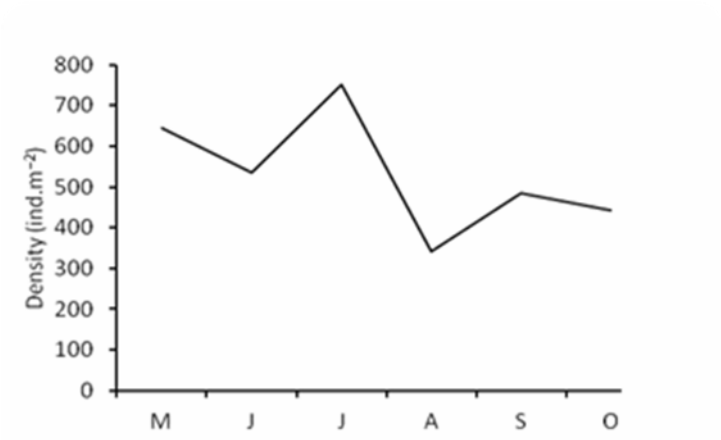


Figure 2. Temporal variation of density of the population of *Idotea balthica* from Agigea.

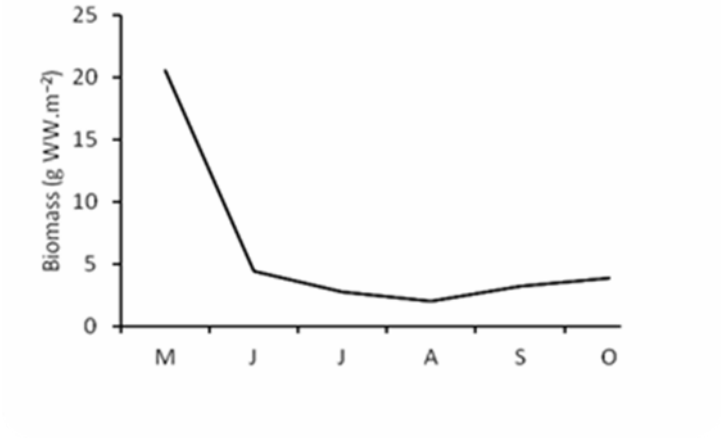


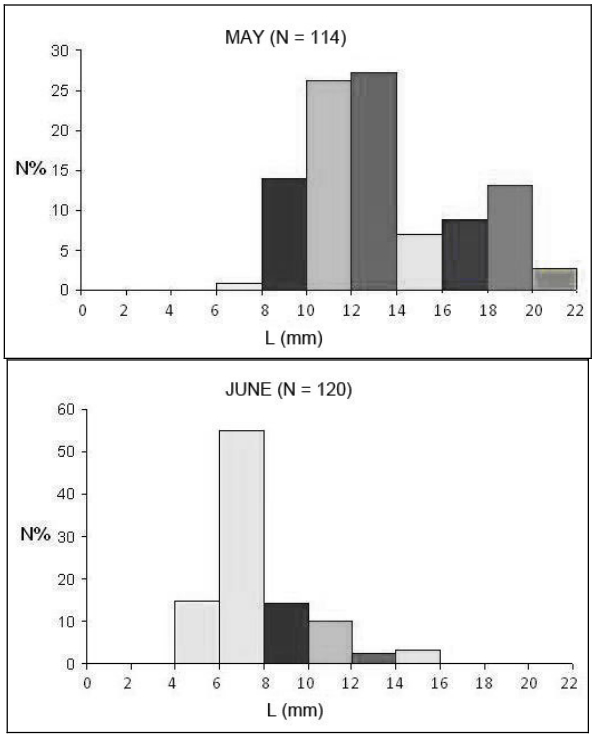
Figure 3. Biomass variation in *Idotea balthica.*

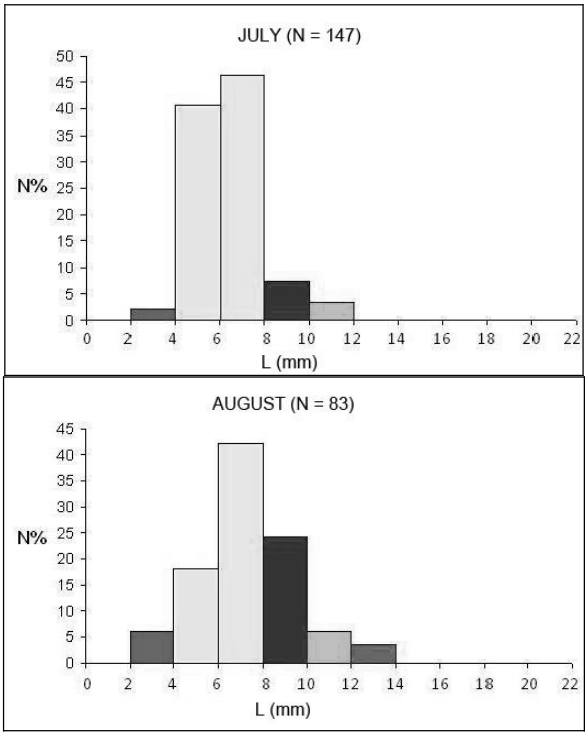
The highest value of biomass, recorded in May (20.4936 g.m^{-2}) is given by adult individuals belonging to class sizes exceeding 8.4 mm. Death of males after reproduction and harvesting of juvenils causes low levels of biomass between June to October, with a minimum in August (1.9467 g.m^{-2}). Biomass sharply decreased in June, but increased the density, which means that individuals collected were mostly juveniles, which can be seen from the analysis of histograms of size classes. From the simultaneous analysis of biomass and density variations graphs results that *Idotea balthica* has two breeding periods: one in May-June and a second one in October. These periods are most likely influenced by the large amount of food due to algal flowering in late May and September.

But, *Idotea balthica* doesn't reproduce in these periods only, it reproduces continuously during the study period, but weaker in July-September. In the samples collected during this period we found pregnant females of *Idotea* wearing a small number of eggs and embryos (minimum values were recorded in July: 4 pregnant females carrying only 63 eggs and embryos).

Measurement of body length of individuals collected made possible a population study by size class, the appearance of cohorts and evolution over time.

In this respect 11 classes of sizes have been established. Length of individuals ranged from 2.88 mm (for individuals in September) and 21.28 mm (for isopodes in May). Charts of May and October are bimodal, indicating the coexistence of two cohorts. For other months, charts are unimodal. In May there were two cohorts (C1 and C2) that disappear in June, replaced by a cohort of juveniles (C3) resulting from reproduction.




JULY (N = 147)
N%
L (mm)
AUGUST (N = 83)
N%
L (mm)

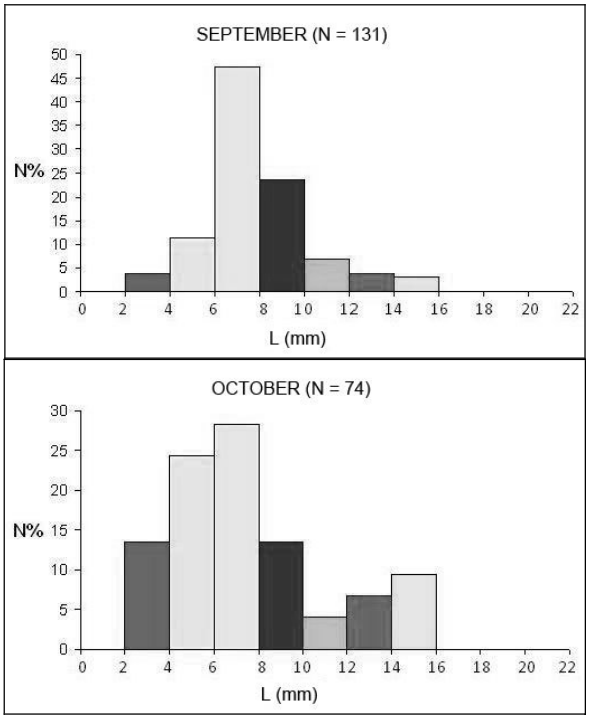


Figure 4. Histograms of the size classes.

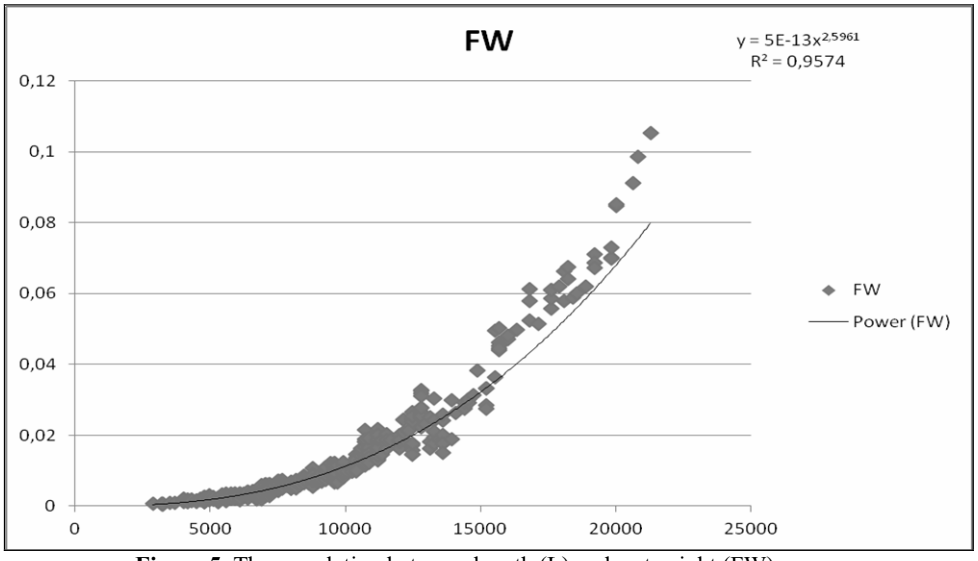


Figure 5. The correlation between length (L) and wetweight (FW).

Idotea balthica is an isomorphic species, which results from the analysis of correlation between the length L and wet biomass FW.

References

Battes, K., Măzăreanu, C., Prıcope, F., Cărăuş, I., Marinescu, V., Rujinschi, R., 2003. *Producţia şi productivitatea ecosistemelor acvatice.* Editura „Ioan Borcea", Bacău.

Boström, C., Mattila, J. 1999. The relative importance of food and shelter for seagrass-associated invertebrates: a latitudinal comparison of habitat choice by isopod grazers. *Oecologia* **120** (1): 162-170.

Boström, C., Mattila, J., 2005. Effects of isopod grazing: an experimental comparison in temperate (*Idotea balthica*, Blatic Sea, Finland) and subtropical (*Erichsonella attenuata*, Gulf of Mexico, U.S.A.) ecosystems. *Crustaceana* **78** (2): 185-200.

Cărăuşu, A., 1955. Contribuţii la studiul isopodelor (*Crustacea Malacostraca)* Mării Negre (litoralul românesc şi regiunile învecinate). III. Familia *Idoteidae*. *Analele Ştiinţifice ale Univ. „Al. I. Cuza" Iaşi*. 137-214.

Dias, N., Hassallb, M., 2005. Food, feeding and growth rates of peracarid macro-decomposers in a Ria Formosa salt marsh, southern Portugal. *Journal of Experimental Marine Biology and Ecology*, **325:** 84– 94.

Downing, J. A., Rıgler, F. H., *A Manual on Methods for the Assessment of Secondary Productivity in Fresh Waters.* Second edition. Blackwell London Publications.

Fowler, J., Cohen, L. 1990. *Practical Statistics for Field Biology*. Open University Press, Milton Keynes.

Gutow, L., Strahl, J., Wiencke, C., Heinz-Dieter Franke, Saborowski, R. 2006. Behavioral and metabolic adaptations of marine isopods to the rafting life style. *Marine Biology* **149**: 821–828.

Honkanen, T., Jormalainen, V., Hemmi, A. 2002. Feeding and growth of the isopod *Idotea baltica* on the brown alga *Fucus vesiculosus*: roles of inter-population and within-plant variation in plant quality. Universitz of Turku, Turku, Finlanda.

Jormalainen, V. 1998. Precopulatory mate guarding in crustaceans - Male competitive strategy and intersexual conflict. *Quarterly Review of Biology*, **73**: 275-304.

Jormalainen, V., Honkanen, T., Heikkilä, N. 2001. Feeding preferences and performance of a marine isopod on seaweed hosts: cost of habitat specialization. *Marine Ecology Progress Series*, **220**: 219–230.

Jormalainen, V., Tuomi, J., Merilaita, S. 1992. Mate choice for male and female size in aquatic isopod *Idotea balthica. Ann. Zool. Fennici*, **29**: 161-167.

Kotta, H. O., Kotta, J., 2004. Food and habitat choice of the isopod *Idotea baltica* in the northeastern Baltic Sea. *Hydrobiologia*, **514**: 79-85.

Muller, G. I., Petran, A., 1976. *Ecologie marină* V. Editura Academiei Republicii Socialiste România. Bucureşti.

Nybbaken, J. W., 1988. *Marine biology: an ecological approach*. 2nd edition. Harper and Row Publishers, New York.

Skolka, M.. *Zoologia nevertebratelor*. (Note de curs). Universitatea „Ovidius" Constanţa.

Surugiu, V., 2005. *Introducere în biologia marină*. Ed. Univ. „Al. I. Cuza" Iaşi.

Surugiu, V., 2008. *Resurse acvatice*. (Note de curs). Universitatea „Alexandru Ioan Cuza" Iaşi.

Tuomi, J., Jormalainen, V., Ilvessalo, H., 1988. Growth, food consumption and reproductive tactics of the aquatic isopod *Idotea baltica. Ann. Zool. Fennici*, **25**: 145-151.

van Der Meer, J., Heip, C. H., Herman, P. J. M., Moens, T., van Oevelen, D.. Measuring the Flow of Energy and Matter in Marine Benthic Animal Pupilations. *Methods for the study of marine benthos.* 326–389.

Vasile, R., 2008. Studiul dinamicii populaţiilor polichetului *Neanthes succinea* din zona canalului Dunare – Marea Neagră. Lucrare de licenţă. Iaşi.

EVOLUTION OF MICROBIAL DENSITY AND BIOMASS IN DIFFERENT GRAIN-SIZED MARINE SEDIMENT MICROCOSMS

DAN RAZVAN POPOVICIU[1] and IOAN ARDELEAN[1,2]
[1] „Ovidius" University of Constanta, Natural Sciences and Agricultural Sciences Faculty, Mamaia Avenue, no. 124, 900527, Constanta, Romania, e-mail: dr_popoviciu@yahoo.com
[2] Biology Institute of Bucharest, Splaiul Independentei, no. 296, 060031 Bucharest, Romania, e-mail: ioan.ardelean57@yahoo.com

Abstract. The influence of sediment granulometry on benthic microbiota and the sediment colonization dynamics were studied in four microcosms containing marine sediments sterilized and sorted according to their grain diameter, and seawater. Their evolution was observed on a two month-period, by sampling every two weeks. Microbial density and biomass were determined by epifluorescence microscopy, using SYBR Green I as a fluorochrome. The sediment colonization process was a rapid one, after two weeks densities reaching 0.77-2.25x10^8 cells.cm^{-3}, and dry biomass reaching 3.6-10 µg.cm^{-3}. Growth continued but at a slower rate. A negative correlation between bacterial cell density and sediment grain size was observed, however not a proportional one.

Key words: bacteria, biomass, marine sediments, granulometry

Rezumat. Evoluția densității şi biomasei microbiene în microcosmosuri cu sedimente marine de diferite granulații. Influența granulometriei sedimentelor asupra microbiotei bentice şi dinamica colonizării acestora au fost studiate în patru microcosmosuri conținând sedimente marine sterilizate şi sortate în funcție de diametrul granulelor, şi apă de mare. Evoluția acestora a fost înregistrată pe parcursul a două luni, prin prelevarea de carote la fiecare două săptămâni. Densitatea şi biomasa microbiană au fost determinate prin intermediul microscopiei de epifluorescență, utilizând SYBR Green I drept fluorocrom. Procesul de colonizare a sedimentelor a fost unul foarte rapid, după primele două săptămâni atingându-se densități de ordinul a 0,77-2,25x10^8 celule.cm^{-3}, respectiv biomase uscate de 3,6-10 µg.cm^{-3}. Creşterea a continuat ulterior, dar într-un ritm mai lent. S-a constatat existența unei corelații negative evidente între densitatea celulelor bacteriene şi diametrul granulelor de sediment, fără ca această corelație să fie una proporțională.

Cuvinte cheie: bacterii, biomasă, sedimente marine, granulometrie

Introduction

From a microbiological point of view, sediments and not the water column are the richest marine environment, marine bacteriobenthos playing an extremely important role in the decomposition of organic matter and nutrient recycling in coastal ecosystems.

Abundance, biomass and composition of sediment microbiota are influenced by various factors, such as sediment granulometry, grain microtopography, water dynamism, oxygenation, protozoan grazing etc., thus inducing geographical and seasonal variations. (Nickels *et al.*, 1981; DeFlaun & Mayer; 1983; Novitsky & MacSween, 1989; Šestanović *et al.*, 2005).

Regarding the relationship between grain size and cell density, there are various opinions. Some studies suggest a significant correlation (Dale, 1974), a partial correlation, dependent on other environmental factors (DeFlaun & Mayer, 1983), or the absence of any predictible relation (Cammen, 1982).

The objective of the current paper was to determine the way bacterial abundance and biomass vary in different grain-sized sediments and the temporal dynamics of bacterial colonization, using sediments from the Romanian Black Sea littoral as a substratum.

Materials and mehtods

Marine sand was collected from several beaches in Constanța, washed with Tween 80 (1 $mg.l^{-1}$ final concentration), dried and sterilized at 105°C and sorted according to grain size using an Endecotts Minor granulometric device, with 1,000, 800, 400, 200, 180, 125, 90 and 53µm sieves.

Sediment was then distributed in four 1 L plastic recipients (filling about one third of each; Fig. 1). Microcosm A contained grains with a diameter between 0-200 µm, microcosm B, 200-400 µm grains, microcosm C, 400-800 µm grains and microcosm D, 800-1,000 µm grains. Seawater (collected from Modern beach) was added to a total volume of around 700 cm^3. Microcosms were covered with transparent foils and kept at room temperature and natural illumination.

Figure 1. Different grain-sized sediment microcosms.

Samples consisting of sediment cores were collected each two weeks, using improvised piston corers (20 ml seringes with the forepart detached, but the gradation intact). From each core, the top 5 cm^3 (coresponding to a 17.5 mm depth) were taken for analysis.

Samples were fixed with buffered formalin (4%; Fry, 1990; Luna *et al.*, 2002) and stored at +4°C.

Dislodgement of bacterial cells attached to sand grains was done by a procedure adapted from existing literature (Epstein & Rossel, 1995; Kuwae & Hosokawa, 1999; Bennett *et al.*, 2006).

Sediment suspensions were diluted 5-fold, incubated with Tween 80 (1 $mg.l^{-1}$ final) for 15 minutes and vortexed at 2,400 r.p.m. for 5 minutes.

Microorganisms were observed by epifluorescence microscopy, using SYBR Green I, an universal fluorochrome that fluoresces bright green when bound to nucleic acids, staining both living and dead cells (Noble & Fuhrman, 1998).

The protocol employed was adapted from Noble & Fuhrman (1998). 2 ml sediment suspensions were diluted to an optimal cell density, incubated for 15 minutes with SYBR Green I (1:10,000 final), filtered on 0.2 µm Millipore filters and examined using a Hund Wetzlar H 600 AFL 50 (1,000 x magnification). An eyepiece grid micrometer was employed.

For each filter, 15-20 grids were randomly chosen, photographed with a digital camera and visualised using MBF ImageJ for Microscopy software (http://www.macbiophotonics.ca/downloads. htm.; Collins, 2007).

Fluorescent cell were manually counted (excluding anorganic and obviously eukaryotic structures, by size and shape) and the average cell density for each sample was determined.

The diameter/length and width of microorganisms were measured using the grid micrometer and biovolume was calculated for each cell (Fry, 1990; Sherr *et al.*, 2001).

To determine dry biomass based on biovolume, several authors proposed different conversion factors. In this study, the following formula was used (Loferer-Krößbacher *et al.*, 1998):

$$m_d = 435 \times V^{0.86}$$

where:
m_d = dry biomass (fg);
V = cell volume (μm^3).

Dry biomass was determined for each cell, then the average value for each sample was calculated.

Results and discussions

After the first two weeks, microbial cell density reached values between 0.77-2.25 × 10^8 cells.cm^{-3} of sediment (Fig. 2). Determinations on freshly sterilized sediments found only 0.03-0.2 × 10^8 cells.cm^{-3} after the treatment described above.

These values fall within the normal range of microbial density in coastal sediments as determined by several authors (for example, Novitsky & MacSween, 1989; Kuwae & Hosokawa, 1999; Luna *et al.*, 2002; Šestanović *et al.*, 2005; Popoviciu & Ardelean, 2011). Growth rate then decreases, with a tendency towards the stabilization of cell density, after 6-10 weeks.

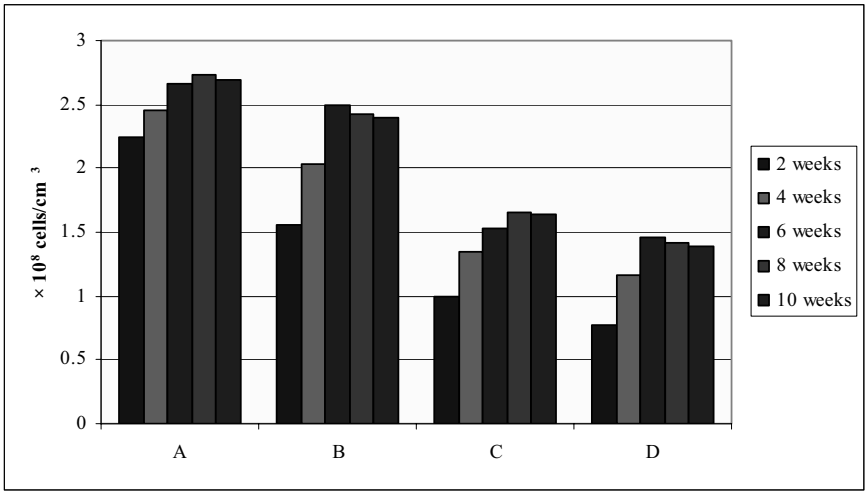


Figure 2. Temporal evolution of microbial density in each microcosm.

Average cell biovolume ranged between 0.069-0.1 μm^3, values situated at the lower limits or immediately below those determined by other researchers (Cammen, 1982; Kuwae & Hosokawa, 1999; Mudryk & Podgórska, 2006; Popoviciu & Ardelean, 2011). Most of the cells observed were bacilli (rod-shaped) and coccobacilli, with cocci accounting for less than one third of the total population, and filamentous forms being rarely found. With similar average biovolumes in all the samples, the temporal evolution of microbial biomass generally followed that of cell density, starting from values betwwen 3.58-10.02 $\mu g.cm^{-3}$ sediment, after the first two weeks (Fig. 3).

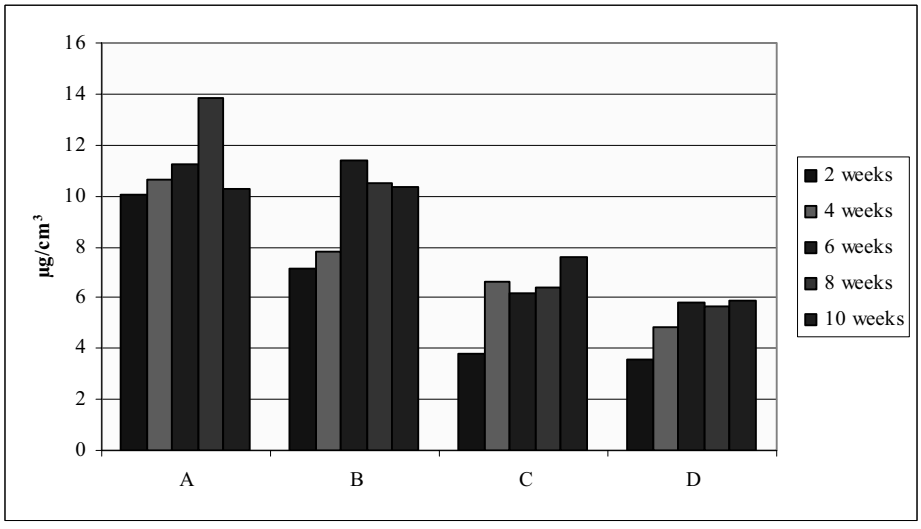
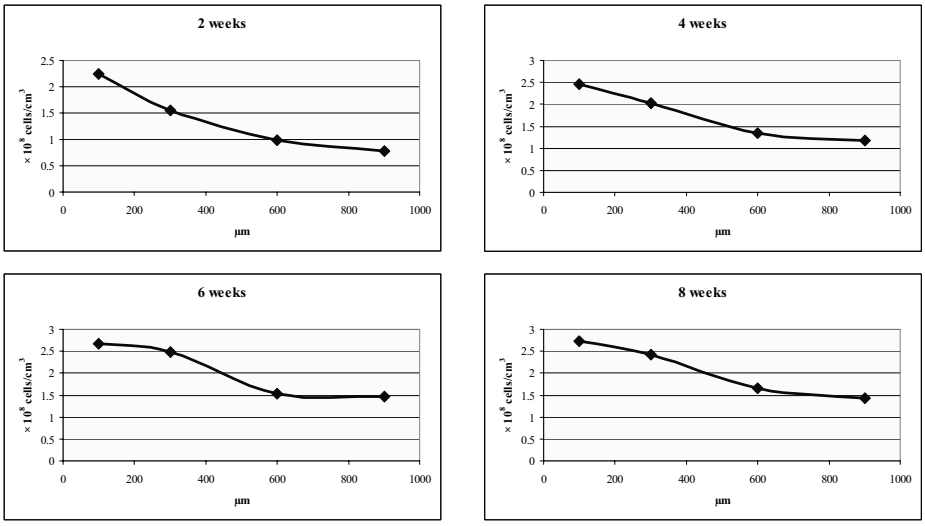


Figure 3. Temporal evolution of bacterial dry biomass in each microcosm.

A relationship between sediment granulometry and the microbiota can be observed. Various researchers had different conclusions regarding this relationship. Dale (1974), studying intertidal sediments, concluded that there was a strong negative correlation between the mean diameter of sediment grains and bacterial abundance (although, theoretically, the distribution of bacteriobenthos should depend on the available surface, the correlation between grain area, mathematically estimated, and cell density was weaker). Nickels *et al.* (1981), experimenting on microcosms and sterilized sediments exposed to seawater discovered the importance of grain microtopography (bacteria colonize mostly surface irregularities). Cammen (1982), in a study on muddy sediments showed the absence of any predictible relation between granulometry and bacterial density, other factors (temperature, grazing) being determinant. On the contrary, DeFlaun & Mayer (1983) noticed a significant (almost linear) dependence of cell density on grain area, but only for a given temperature, a modification of water temperature changing the slope.

Various researches on natural sediments showed a relationship between grain diameter and the characteristics of benthic microbiota, without a mathematical expression (for example, Kuwae & Hosokawa, 1999; Hubas *et al.*, 2006). A study on sandy sediments on the Romanian littoral (Popoviciu & Ardelean, 2011) resulted in significant differences between different grain-sized sands, but without a proportional, nor an absolute correlation, suggesting that local enviromental factors can be at least as important as granulometry.

In the present experiment, under controlled environmental conditions (similar temperatures, lack of water dynamism), a negative correlation between mean grain diameter and cell density was obvious (mean cell biovolume varied independently from granulometry). This correlation was not a proportional one and could not be expressed by any mathematical ecuation (Fig. 4; a possible reason being the irregular shape of sand grains).



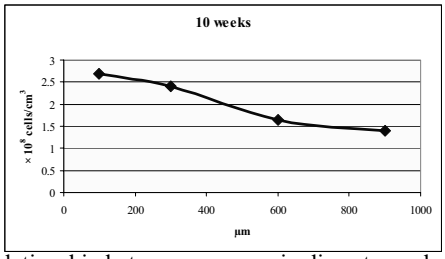


Figure 4. Relationship between mean grain diameter and microbial density.

It should be noted that direct enumeration of bacteria has some limitations. The dislodgement method employed, background fluorescence, very small size of some cells etc., can influence the results (Fry, 1990; Kepner & Pratt, 1994; Gough & Stahl, 2003). The fluorochrome used, the conversion factor, and the presence of some disk-shaped cells can affect biomass determination (Sherr *et al.*, 2001, Mudryk & Podgórska, 2006).

Conclusions

The current paper shows that, under controlled environmental conditions, microbial abundance is correlated to the mean diameter of sediment particles. An increase in grain size leads to lower cell densities. However, this correlation is not a proportional one.

Mean cell biovolume independently varied from granulometry.

Microbial colonization of sterile sediments was a very rapid process, in only two weeks cell densities reaching values similar to those in natural sediment samples. This might suggest that a large part of marine sediment microbiota has planktonic origins.

Acknowledgements

Special thanks to conf. dr. Teodora Onciu, for technical support in granulometric sorting.

References

Bennett, P. C., Engel, A. S., Roberts, J. A., 2006. Counting and imaging bacteria on mineral surfaces. In Patricia, J., Maurice, A., Warren, L.A. (eds.) Methods of Investigating Microbial-Mineral Interactions. *CMS Workshop Lectures*, **14**: 37-78, The Clay Mineral Society, Chantilly.

Cammen, L. M., 1982. Effect of particle size on organic content and microbial abundance within four marine sediments. *Mar. Ecol. Prog. Ser*., **9**: 273-280.

Collins, T. J., 2007. Image for microscopy. *BioTechniques*, **43**: 25-30.

Dale, N. G., 1974. Bacteria in intertidal sediments: factors related to their distribution. *Limnol. Oceanogr*., **19**: 509-518.

DeFlaun, M. F., Mayer, L. M., 1983. Relationships between bacteria and grain surfaces in intertidal sediments. *Limnol. Oceanogr.*, **28**: 873-881.

Epstein, S. S., Rossel, J., 1995. Enumeration of sandy sediment bacteria: search for optimal protocol. *Mar. Ecol. Prog. Ser.,* **117**: 289-298.

Fry, J. C., 1990. Direct methods and biomass estimation. *Meth. Microbiol.*, **22**: 41-85.

Gough, H. L., Stahl, D. A., 2003. Optimization of direct cell counting in sediment. *J. Microbiol. Meth*., **52**:
39-46.

Hubas, C., Davoult, D., Cariou, T., Artigas, L. F., 2006. Factors controlling benthic metabolism during low tide along a granulometric gradient in an intertidal bay (Roscoff Aber Bay, France). *Mar. Ecol. Prog. Ser*., **316**: 53-68.

Kepner, R. L., Pratt, J.R., 1994. Use of fluorochromes for direct enumeration of total bacteria in environmental samples: past and present. *Microbiol. Rev.*, **58**: 603-615.

Kuwae, T., Hosokawa, Y., 1999. Determination of abundance and biovolume of bacteria in sediments by dual staining with 4',6-diamidino-2-phenylindole and acridine orange: relationship to dispersion treatment and sediment characteristics. *Appl. Environ. Microbiol.,* **65**: 3407-3412.

Loferer-Krößbacher, M., Klima, J., Psenner, R., 1998. Determination of bacterial cell dry mass by transmission electron microscopy and densitometric image analysis. *Appl. Environ. Microbiol*., **64**: 688-694.

Luna, G. M., Manini, E., Danovaro, R., 2002. Large fraction of dead and inactive bacteria in coastal marine sediments: comparison of protocols for determination and ecological significance. *Appl. Environ. Microbiol*., **68**: 3509-3513.

Mudryk, Z. J., Podgórska, B., 2006. Scanning electron microscopy investigation of bacterial colonization of marine beach sand grains. *Baltic Coastal Zone*, **10**: 61-72.

Nickels, J. S., Bobbie, R. J., Martz, R. F., Smith, G. A., White, D. C., Richards, N. L., 1981. Effect of silicate grain shape, structure, and location on the biomass and community structure of colonizing marine microbiota. *Appl. Environ. Microbiol*., **41**: 1262-1268.

Noble, R. T., Fuhrman, J. A., 1998. Use of SYBR Green I for rapid epifluorescence counts of marine viruses and bacteria. *Aquat. Microb. Ecol.,* **14**: 113-118.

Novitsky, J. A., MacSween, M. C., 1989. Microbiology of a high energy beach sediment: evidence for an active and growing community. *Mar. Ecol. Prog. Ser.,* **52**: 71-75.

Popoviciu, D. R., Ardelean, I. I., 2011. Bacterial abundance, biomass and proportion of living cells in littoral sandy sediments on the Romanian Black Sea coast. *SGEM* 2011, **2**: 995-1002.

Sherr, B., Sherr E., Del Giorgio, P., 2001. Enumeration of total and highly active bacteria. *Meth. Microbiol.*, **30**: 129-159.

Šestanović, S., Solić, M., Krustulović, N., Bogner, D., 2005. Volume, abundance and biomass of sediment bacteria in the eastern mid Adriatic Sea. *Acta Adriat.*, **46**: 177-191.

SEVERAL CHARACTERISTICS OF THE MOUFLON POPULATION (*OVIS AMMON MUSSIMON* PALLAS, 1811) FROM THE MUSEUM COMPLEX OF NATURAL SCIENCES OF CONSTANTA (C.M.S.N.)

VERONICA ANTONE
Museum Complex of Natural Science, Bd. Mamaia no. 255, Constanta, Romania
e-mail: veronica.antone@gmail.com

Abstract. Starting from 1987, were brought to "Microreserve" – section of C.M.S.N. Constanta, 3 mouflon specimens (2 males, 1 female) (*Ovis ammon mussimon* Pallas 1811) from the hunting fond of Negureni, which belongs to the District of Forest Baneasa. By analyzing 12 somatometric parameters of 20 specimens with ages between 1 and 10 years, it was found a slightly decrease over time of body dimensions, yet the average values are remaining close to those found in literature. This work presents the dynamics of mouflon's population beginning with 1987 until the present day, meaning 23 years of research.

Key words: mouflon, population dynamics, database, software

Rezumat. Câteva caracteristici ale populației de mufloni din cadrul Complexului Muzeal de Ştiinte ale Naturii - Constanța. Începând din anul 1987, în cadrul „Microrezervației" - secție a C.M.S.N. Constanța, au fost aduse 3 exemplare de mufloni (2 masculi, 1 femelă) (*Ovis ammon mussimon* Pallas 1811) din fondul cinegetic Negureni, aparținând Ocolului Silvic Băneasa. Analizând 12 parametri somatometrici la 20 exemplare cu vârste între 1 şi 10 ani, s-a constatat o uşoară scădere a dimensiunilor corporale în timp, mediile valorilor rămânând însă apropiate de cele din literatură. Lucrarea de față prezintă şi dinamica populației de mufloni din anul 1987 până în prezent, reprezentând 23 de ani de cercetare.

Cuvinte cheie: muflon, dinamica populației, bază de date, software

Introduction

The existing records shows that in Romania the European wild sheep colonization, brought from the islands Sardinia (Italy) and Corsica (France), has been tried for several times, but only in the region of Dobrogea the climate permitted an optimal evolution of the mouflon populations.

Researches made in 1961 and 1962 by the laboratory of Forest Research and Management Institute Bucharest (I.C.A.S.) recommended that the mouflon colonization to be made in different locations across Romania and among these locations was Dobrogea.

In 1965 and 1966 begins the mouflon colonization into the forests of Vlahi (Forest District Cernavoda) and Negureni (Forest District Baneasa) and in 1971, into the M. Eminescu Forest (Forest District Baneasa).

According to the C.M.S.N. records, the first mouflons were 2 males and 1 female, and they were brought form the Valea Cismelelor Forest, Negureni village - District of Forest Baneasa, Constanta County, and they were included in the patrimony of C.M.S.N. in August 8, 1987. They originate from the breeding of mouflons brought from Vienna in 1966, at District of Forest Baneasa (Geacu *et al.*, 2007).

Opened in 1985, "Microreserve" is part of the Museum of Natural Sciences Constanta. It is located in the proximity of holiday resort Mamaia. It covers a surface close to 6 ha, with 2 ha of watershed originated in Tabacarie Lake.

The mouflon (*Ovis ammon mussimon* Pallas, 1811) belongs to order Artiodactyla Owen, 1848, the family of Bovidae (Graz, 1821) and genus *Ovis* Linnaeus, 1758.

Material and methods

The study covers the interval between 1987 and 2011; 4 years are representing personal studies.

Concerning the mouflon population from CMSN, the following aspects have been observed and analyzed:

- Quantitative evolution: populations dynamic's, birth rate, mortality rate;
- Qualitative evolution: somatometric data, horns' growth rhythm, fertility;
- Comparative study: using data obtained from other zoological gardens and hunting parks from Romania.

The data concerning the period between 1987 and 2007 were extracted from the C.M.S.N. records.

Data concerning the mouflon characteristics and the mouflon population, for the interval between 2008 and 2011, were acquired by following stages:

- Individual identification (by using RFID tags)
- Age determination
- Biometric characterization
- Weight measurement
- Observations over time

Age determination for the mouflon was made by using the method of ovine age determination, based on dentition (Voia, 2005). Concerning the males, it was taken into account the number of yearly circles of their horns (Cota & Bodea, 1969).

The somatometric data was obtained by measuring 12 parameters over well defined time intervals: at 6 months for mature individual, at 1 month for juvenile and at 1 week for offspring with age between 1 and 60 days.

The measured body parameters are presented into the table 1. The followings were under consideration: 3 lengths, 3 heights, 2 widths, 2 perimeters, 1 depth and 1 weight.

Table 1. The measured body parameters.

No	Specification	Parameter
1	lengths =3	total length
2		body length
3		tail length
4	heights =3	shoulder height
5		loin height
6		ear height
7	widths = 2	shoulder width
8		loin width
9	perimeters =2	thorax perimeter
10		lower leg perimeter
11	depths=1	thorax depth

12	weight=1	liveweight

Corporal measurements were made by zoometric compass and tapemeasure.

For the determination of horn's growth rate by age groups, 10 mouflon males were observed, with ages between 1 and 10 years.

The natality and mortality rates were calculated with formulas elaborated by Uloth, 1963, and Ilie, 1975.

Comparative analysis was made by using a custom database software application. The application has a client-server design, built upon MySQL database server database, and having browser-based user interface for friendly datamining and reporting. The main goal of the application is to create a colaborative framework. The database contains data which describe the mouflon nucleus from Romania (Fig. 1), somatometric data (Fig. 2), periodical notes or recordings.

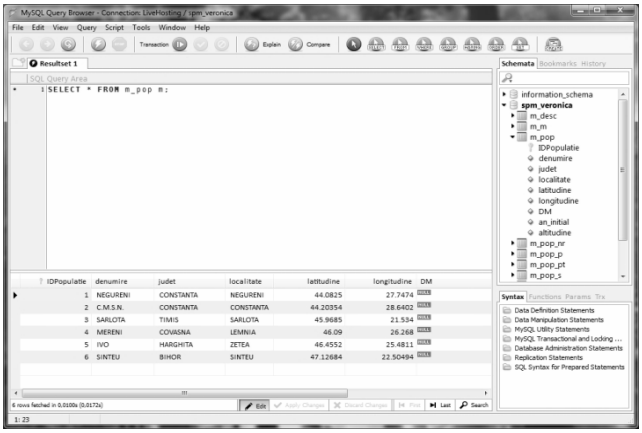


Figure 1. Screen capture of typical table view.

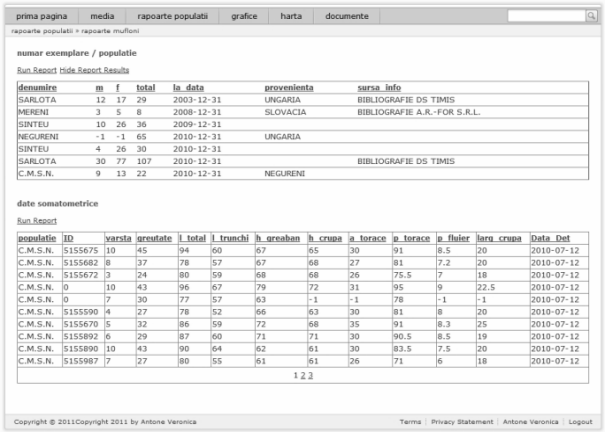


Figure 2. Screen capture of application user interface.

The observations were made by simple inspection and have been documented by using a photo camera Fujitsu FinePix S1000fd.

Results and discussions

Mouflon characteristics

Analyzing the acquired data, we may conclude the following:

The average values of the 12 measurements are higher at males than females, but we have to remark that the lengths differs in lower limits; the differences are higher considering heights, perimeters and liveweights of the specimens (Fig. 2).

The mouflon has permanent horns, continually growing and representing hunting trophies; their value is calculated by length, thickness, color and aspect of the horns.

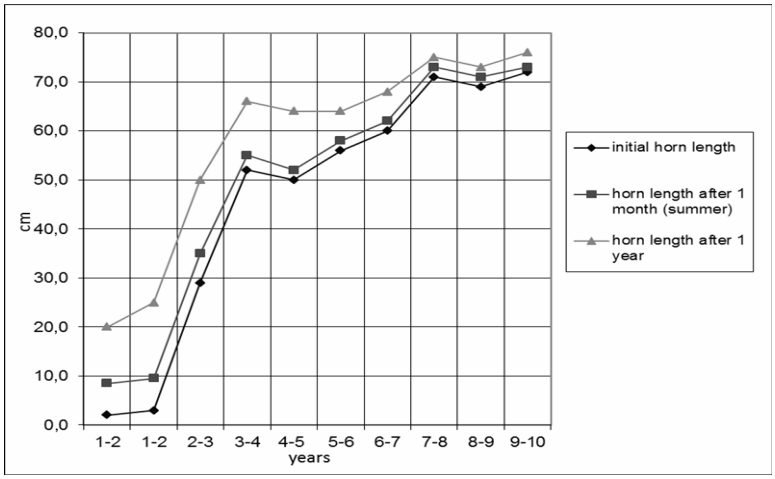


Figure 3. Horn growth rhythm.

Mouflon population characteristics

The mouflon adapted well to the local conditions from "Microreserve". Starting from 3 individuals (2 males and 1 female) in 1987, this small group had continuously grown. In 5 years, this group has grown by 3 times.

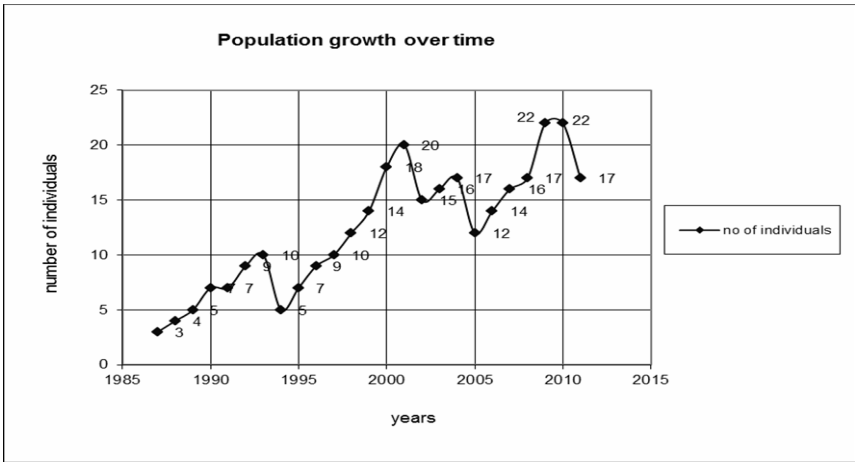


Figure 4. The evolution of mouflon population from C.M.S.N., between 1987 and 2011.

In 1994 and between 2000 and 2005, 26 individuals had been extracted from this group (10 males and 16 females) to populate other zoological gardens across the country (Calarasi, Craiova, and Targoviste), but in spite that fact, the population attained 2 peaks: in 2001 - 20 mouflons and in 2009 - 22 mouflons.

The birth rates are high, and close to other mouflon populations from Europe. In 22 years of study it was observed that in most cases the number of born females was higher or at least equal with the number of males, with only 2 exceptions which occurred in 2006 and 2007.

In the first 7 years, the birth rate has attained a maximum of 100% - reported at females' number. Starting with 1999, due to inbreeding, the birth rate is constant, but the mortality of the lambs is increasing in the first 8 months, and thus the overall growing rate of the population is between 60 and 70%. Starting with 2005, the population growth rate is below 20%.

Mortality rate is below the birth rate. The main causes of mortality are: birth complications, infestation caused by *Echinococcus* spp., gastroenteritis, wandering dog attacks, pulmonary congestion, due to ageing and male fights.

Table 2. Age distribution of the C.M.S.N. mouflon population in 2010.

Gender	**0-1**	**1-2**	**2-3**	**3-4**	**4-5**	**5-6**	**6-7**	**7-8**	**8-9**	**9-10**	**>10**	Grand total
Male	2	0	1	1	1	1	1	1	1	1	0	10
Female	3	1	1	2	1	1	2	0	1	0	0	12
Total	5	1	2	3	2	2	3	1	2	1	0	22
Percent (%)	22.7	4.5	9.1	13.6	9.1	9.1	13.6	4.5	9.1	4.5	0	100%

The sex ratio is normal, with values close to 1/1.

In the table 1 it could be observed a uniform representation of all age groups, with a bigger number of individuals in the group 0-1 years.

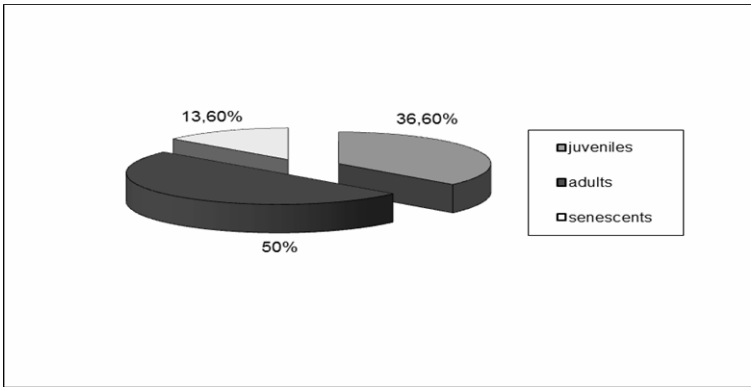


Figure 5. Age groups distribution of mouflon population from C.M.S.N. in 2010.

The real curve compared with the aspect of the ideal curve of a population in progress (Botnariuc & Vădineanu, 1982, after Odum) shows us that the population is in a state of stationary equilibrium. Females are better represented in the juvenile group and in the adult group, but males are those with longer life duration, being represented in age groups bigger than 7 years.

At the end of 2010, there are 317 mouflons in Romania. 229 specimens (63 males, 166 females) are in hunting parks or complexes, and the remaining 88 specimens (41 males, 47 females) are in zoological gardens (Figs. 6-7).

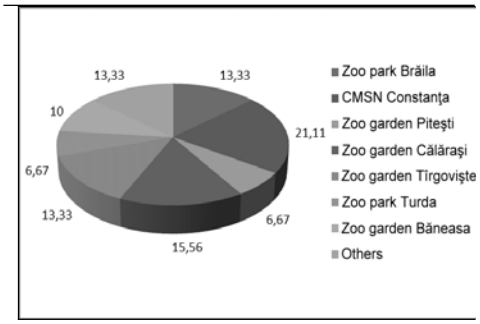


Figure 6. Distribution per zoological garden.

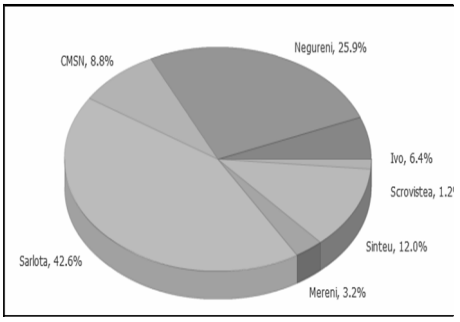


Figure 7. Distribution per population.

Conclusions

The mouflon population from C.M.S.N. Constanta is largest than any other zoological garden from Romania. From this group were extracted 26 mouflons for the enrichment of other zoological gardens' live patrimony.

The evolution of the mouflon population considered shows the breed's degree of acclimatization and the effects of inbreeding due to limited space, climatic conditions and the small number of specimens (three) of the initial population.

For maintaining a higher birth rate and to reduce the influence of consanguinity, in 2010 it was started an exchange program with interested forest districts.

References:

Botnariuc, N., Vădineanu, A., 1982. *Ecologie*. Ed. didactică şi pedagogică, Bucuresti.

Cota, V., Bodea, M., 1969. *Vânatul României*. Ed. Agrosilvică, Bucureşti.

Geacu, S., Crăciun, F., Vintilă, A., 2008. Populația de mufloni (*Ovis ammon* L.) de la Negureni-Băneasa (jud. Constanța). *Revista Geografică*, t. XIV-XV: 55-63.

Ilie, E., 1975. *Aclimatizarea marmotei şi muflonului. Recomandări pentru producție în silvicultură*, Bucureşti.

Uloth, W., 1963. Zur Frage der Misch Olttigkeit beim Muffelwild, Z.f.Jagdwissen schaft 9 (3), Hamburg-Berlin: 112-115.

Voia, O., 2005. *Ovine şi Caprine - Ghid practic de creştere*. Editura Waldpress, Timişoara.

CONTRIBUTIONS TO KNOWLEDGE OF STRUCTURAL DYNAMICS OF THE POPULATIONS OF MEDICINAL LEECHES IN THE BRAILA MARSHES COMPLEX / THE LOWER DANUBE WETLAND SYSTEM

TIBERIU ANDREI FERMAS and SERGIU CRISTOFOR
Department of Systems Ecology and Sustainability, University of Bucharest, Bucharest, Romania,
e-mail: tiberiu_fermas@yahoo.com

Abstract. The populations of genus *Hirudo*, in the aquatic ecosystem of the Small Island of Braila, have been less investigated despite the fact they represent a component with an important abundance of the community and that the medicinal leeches are appreciated for the services they offer. The researches on the medicinal leeches in Romania are sporadic and reduced to some taxonomic and anatomical aspects that have been achieved in the twentieth century. This paper present the first results of a research programme that aims to develop knowledge on structural and functional dynamics of the populations of genus *Hirudo* (Fam. *Hirudinidae*) in the Braila Marshes complex (Ramsar Site and natural parc). The research programme is extended for three years (2010 - 2012) and aims at: i) identifying spatial-temporal complexity of organizations and functioning of these populations, ii) differentiating the major natural and anthropogenic driving forces and pressure factors responsible for their dynamics and decline, iii) modeling the dynamics of these populations, identified as heterogeneous units with complex structure and iv) providing scientific information for sustainable use of these resources. Preliminary analysis of samples taken during the first year of research indicates the presence of the species *Hirudo verbana* (Carena).

Key words: medicinal leeches, *Hirudo verbana, Hirudo medicinalis*, Romania, Small Island of Braila

Rezumat. Contribuţii la cunoşterea dinamicii structurale a populaţiilor de hirudinee medicinale din complexul Bălţile Brăilei / Sistemul de Zone Umede al Dunării Inferioare. Populaţiile genului *Hirudo* din ecositemele acvatice ale Bălţilor Brăilei au fost mai puţin cercetate în ciuda faptului că reprezintă o componentă cu abundenţă importantă a biocenozelor, lipitorile medicinale prezentând interes pentru serviciile oferite. Cercetările asupra lipitorilor medicinale din România sunt sporadice şi reduse asupra unor aspecte taxonomice şi anatomice realizate în secolul al XX-lea. Lucrarea de faţă prezintă primele rezultate ale unui program de cercetare ce îşi propune dezvoltarea cunoaşterii asupra dinamicii structurale şi funcţionale a populaţiilor genului *Hirudo* (fam. *Hirudinidae*) din complexul Bălţile Mici ale Brăilei (sit Ramsar şi Parc Natural). Programul de cercetare este extins pe trei ani (2010-2012) şi îşi propune să contribuie la: i) identificarea complexităţii organizării spaţio-temporale şi a funcţionării acestor populaţii; ii) diferenţierea principalilor factori de comandă naturali şi antropici şi a căilor de exercitare a presiunii, care au determinat şi determină dinamica, respectiv dec1inul acestora; iii) modelarea dinamicii acestor populaţii, identificate ca unităţi heterogene, cu structură complexă şi iv) fundamentarea ştiinţifică a utilizării durabile a acestor resurse. Analizele preliminare asupra probelor prelevate în cursul primului an de studiu indică prezenţa speciei *Hirudo verbana* (Carena).

Cuvinte cheie: lipitori medicinale, *Hirudo verbana, Hirudo medicinalis*, Romania, Insula Mică a Brăilei

Introduction

Although medicinal leeches have been used since ancient times to treat various medical affections (Whitaker *et al.*, 2004, Codell, 2001) there is a lack of information regarding ecology and status within the natural ecological systems. The main results are provided from laboratory studies and cannot be applied to natural populations (Demirsoy, 2001). At national level, the information about the ecology and biology of medicinal leeches is scanty (Scriban, 1904, 1910; Radu and Radu,

1972; Fira and Nastasescu, 1977). Also there is a lack of studies regarding the national hirudofauna, which is due to the low interest for this species rather than to its absence (Utevsky *et al.*, 2010).

To stop the exploitation of the natural populations by collecting (Sawyer, 1981) and low down the possibility of extinction, the medicinal leech *Hirudo medicinalis* is protected through international (Red List – IUCN Appendix II of the CITES, Berne Convention, Annex V on the EU Council directive 92/43/EEC on the conservation of the natural habitats and of wild fauna and flora) and national legislation measures.

The main objective of this material is to clarify the existence of medicinal leeches in the research area and to contribute to the actual stage of knowledge with new information about the composition and the structure of the medicinal leeches populations.

Description of sites studied

The Lower Danube Wetland System (LDWS) comprises five major hydrogeomorphological units. One of them is represented by the Inner Danube Delta which has developed on the Romanian territory along the river stretch between Calarasi (365 km) and Braila (170km) and between Southern Romanian Plain and the Dobrogean Plateau over a total surface of 2413 square kilometers. The Inner Danube Delta has as main components the Small and Big Islands of Braila (176, respectively 700 square kilometers), the Borcea Island (801 square kilometers) and the lateral flooding areas (736 square kilometers) (Vadineanu, 2001).

The Small Island of Braila - SIB (Ramsar site, Natural Park and international socio-ecological long-term research area) is characterized by a natural flooding regime which during the evolution gave birth to a number of 10 islands and channels that comprise 13 types of habitats or categories of ecosystems. Among these more than 50% are natural ecosystems, about 30% are currently semi-natural ecosystems and less than 20% have been affected by direct human intervention. The richness of fauna identified in SIB is represented by 136 species of birds (47 species listed on the annex of EU-Bird Directive and 34 species on the annex of Bern Convention), 11 species of mammals, 13 species of amphibians and reptiles, 65 species of fish, 49 higher taxa of terrestrial invertebrates, over 100 species of Gastropoda and Bivalvia and more than 60 species of benthic organisms (Vadineanu *et al.*, 2001).

The research was conducted in two complexes of aquatic ecosystems representative for the SIB (Fundu Mare, Popa) and one lake for the lateral flooding areas (Piatra Fetei). In Fundu Mare complex the research was focused on the lakes Chiriloaia si Bordeiele and in Popa complex the sample units have been established in the lakes Gasca, Curcubeu and Vulpasu (Fig. 1).

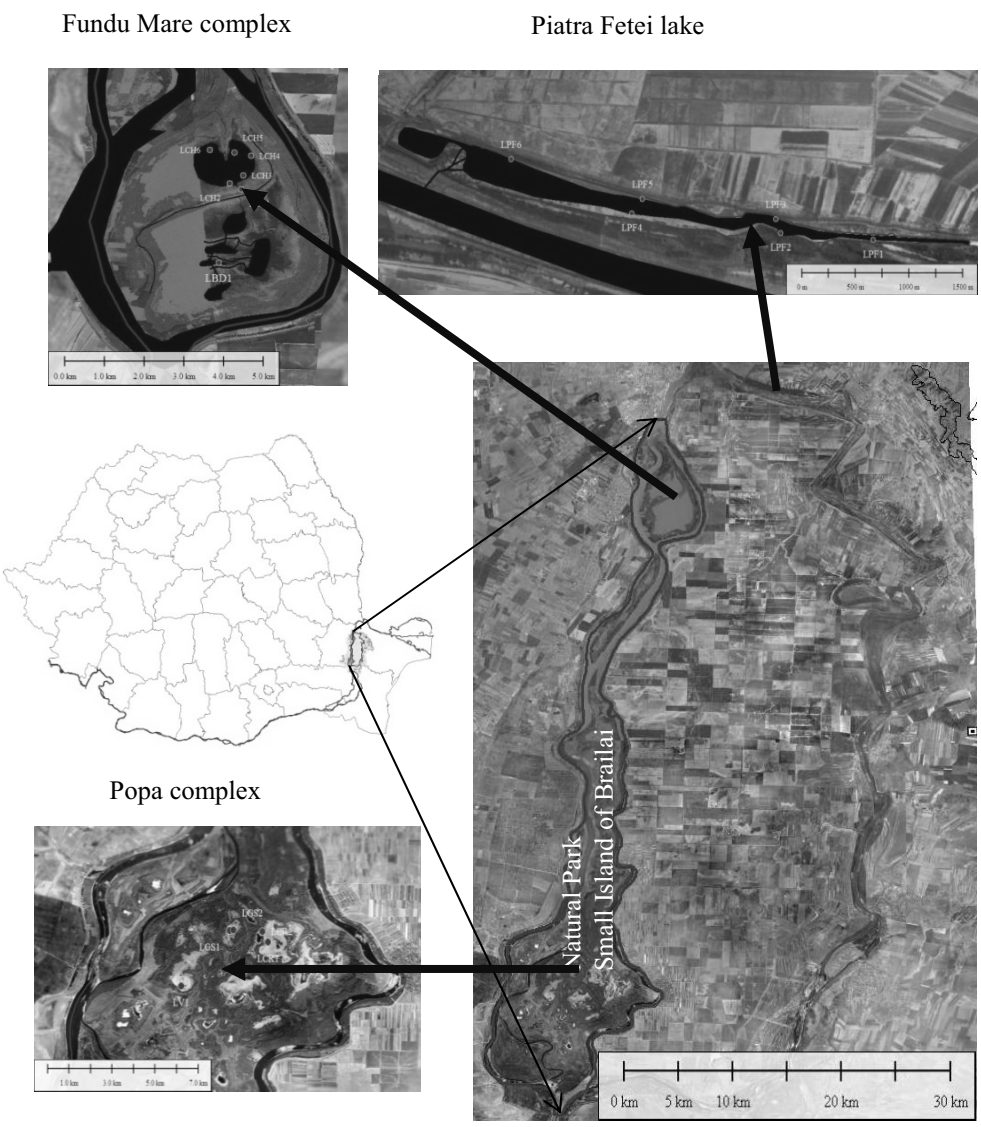


Figure 1. Location of the study sites and placement of the stations.

Materials and methods

The medicinal leeches have been collected during the warm season (Mai-September 2011), in the first week of each calendar month. In each lake, in different areas, there have been established sampling stations, depending on the hydro-connectivity and specific elements for medicinal leeches (water depth, shading degree, presence of amphibians and vegetation) formed of a sample unit. Depending

on the surface of the body of water but also on its depth the sampling stations have been adjusted from one month to the other. In total there have been established 46 sampling stations.

The sampling method used is the one recommended by Elliott and Tullett, 1986 and consists of disturbing the water by an operator in a known period of time. The period of water disturbing was of 40 minutes. Disturbing the water led to attracting medicinal leeches, which have been sampled by hand or by small eyes fish landing. The catch-per-unit-effort was therefore the total number of medicinal leeches taken by one operator in 40 minutes. In the area of the established sampling stations, the medicinal leeches have been also sampled from the specific hiding places (under rocks, in vegetable waste), when the case.

After sampling, the leeches have been introduced in bottles with water and weighted the same or the next day in the laboratory with an electronic balance to the nearest 0.01 g. Before weighting, to discard the water surplus the leeches have been tamponed with a cotton material.

Results

The presence of two species of leeches has been identified in the research area: *Hameopis sanguisuga* and the medicinal leech *Hirudo verbana* (Fig. 2).

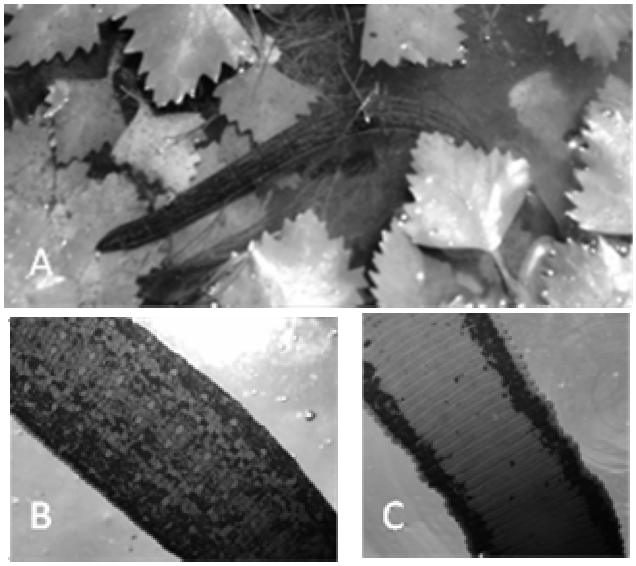


Figure 2. *Hirudo verbana* sp. – external description - (A, B - dorsum, C - venter).

In total there have been sampled 1139 *Hirudo verbana* leeches. Within the researched complexes the greatest number of sampled leeches has been from Chiriloaia (Mai) and the smallest from Piatra Fetei (September). The distribution of the total number of leeches and of the sampling stations within each researched aquatic complex is shown in Table 1.

Table 1. Dynamics of total number of stations and sampled leeches (N) in Fundu Mare, Popa, Piatra Fetei in Mai-September 2011.

	Mai		June		July		August		September		Total no. of stations	Total N
	No. of stations	N	No. of stations	N	No. of stations	N	No. of stations	N	No. of stations	N		
Fundu Mare	6	369	5	98	1	195	5	115	-	-	17	777
Popa	-	-	-	-	5	274	2	11	-	-	7	285
Piatra Fetei	5	21	6	29	5	16	5	9	1	2	22	77
Total	11	390	11	127	11	485	12	135	1	2	46	1139

The dynamics of numeric density and biomass presented differentiated characteristics in time (different moments of active life) and space (from one lake to the other or heterogeneousness within the same lake). The total number of sampled individuals in Chiriloaia fluctuated very much on the whole period of the research. Numerical abundance in Piatra Fetei is very different to the one in Chiriloaia (Fig. 3).

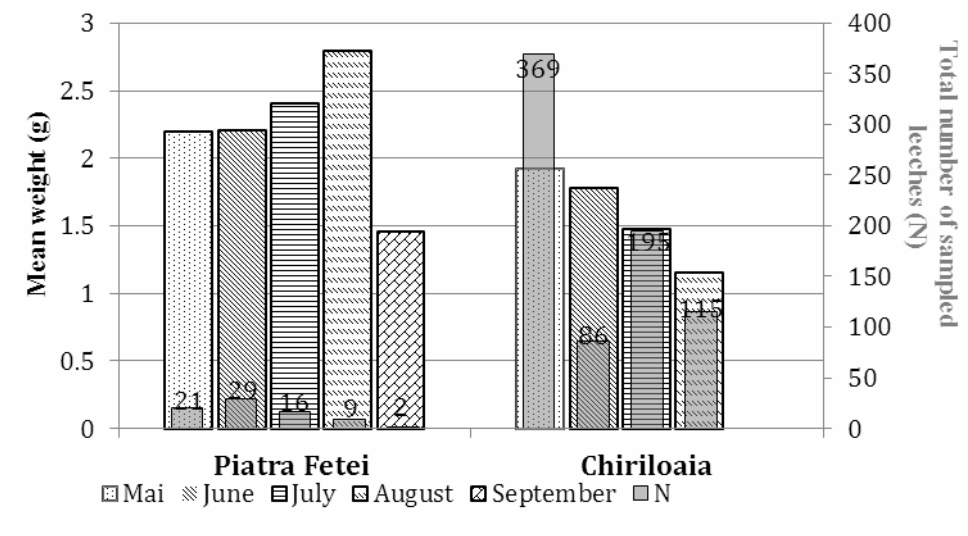


Figure 3. Dynamics of numerical density and mean biomass in Piatra Fetei and Chiriloaia, Mai-September 2011.

During the research, the numerical density of *Hirudo verbana* was high in Chiriloaia, Gasca and Vulpasu. The number of medicinal leeches differed from one lake to another, the exception being Curcubeu and Vulpasu in July, with a mean number of sampled individuals of 80.5, respectively 80. Also the mean biomass registered in the two lakes had similar values (Fig. 4). Although the lakes Gasca and

Curcubeu communicate through interior channels, a significant difference in wet biomass but also in the number of sampled leeches has been registered. The smallest number of sampled leeches during the whole period of sampling has been registered in Piatra Fetei, Gasca and Bordeiele (Fig. 3-4).

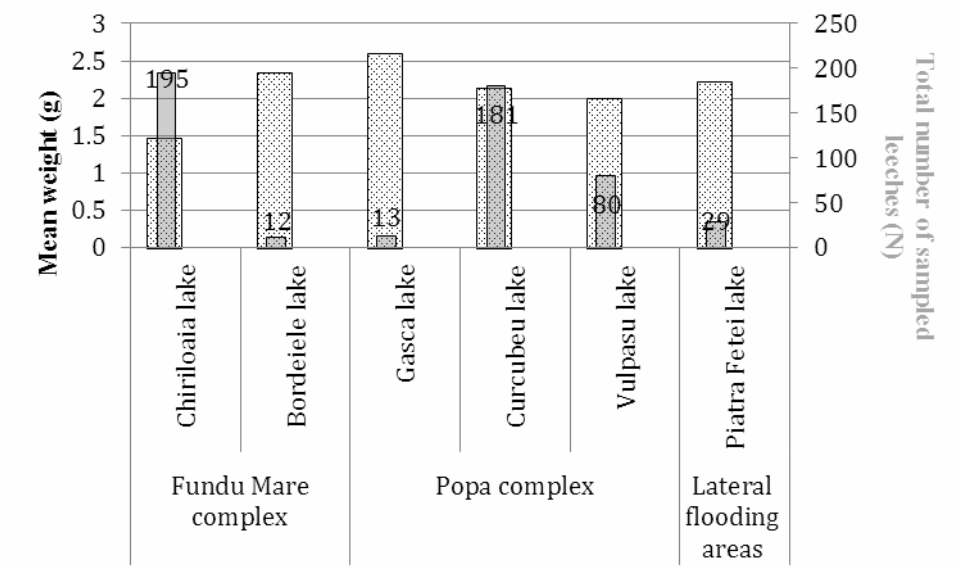


Figure 4. Dynamics of numerical density and mean biomass in Fundu Mare, Popa and Piatra Fetei, July 2011.

The mean biomass decreases constantly in Mai-August, the smallest value being registered in August (1.15 g); also the smallest weight (0.02 g) has been registered in June (Chiriloaia). The highest mean biomass (2.80 g) and the highest absolute minimum (2 g) have been registered in Piatra Fetei in August (Table 2).

Table 2. Monthly mean, minimum and maximum biomass of leeches sampled from Piatra Fetei and Chiriloaia Mai-September 2011.

	Piatra Fetei			Chiriloaia		
	Mean (g)	Min (g)	Max (g)	Mean (g)	Min (g)	Max (g)
May	2.20	0.67	6.45	1.93	0.30	9.37
June	2.20	0.97	5.41	1.79	**0.02**	5.67
July	2.40	1.15	4.25	1.47	0.30	5.86
August	**2.80**	**2.00**	4.35	**1.15**	0.06	3.06
September	1.46	1.31	1.60	-	-	-

In the study area, during the whole period of research, the highest value of weight registered was of 9.51 g. in Curcubeu in July (Table 3).

Table 3. Monthly mean, minimum and maximum biomass of leeches sampled in Bordeiele, Curcubeu, Gasca si Vulpasu, July-August 2011.

	Bordeiele			Curcubeu			Gasca			Vulpasu		
	Mean (g)	Min (g)	Max (g)	Mean (g)	Min (g)	Max (g)	Mean (g)	Min (g)	Max (g)	Mean (g)	Min (g)	Max (g)
July	2.33	0.56	7.27	2.15	0.19	**9.51**	2.61	1.31	6.12	1.9	0.33	4.97
Aug	-	-	-	-	-	-	2.04	1.07	3.37	-	-	-

Discussion

Differentiating the species of *Hirudo* genus by methods which use morphological (Utevsky and Trontelj, 2005) and genetic aspects (Trontelj *et al.*, 2004, Trontelj and Utevsky, 2005; Utevsky and Trontelj, 2005, *Siddall et al.*, 2007) eliminates anterior confusions (*Hirudo verbana* was assigned to *Hirudo medicinalis*) and allows correct establishment of species. The medicinal leech identified in the research area is *Hirudo verbana*.

The presence of this species on Romanian territory was uncertain at international level and the first mention about its existence at national level has been recently conducted (Gagiu, 2010). The confirmation of the presence of the species *Hirudo verbana* on Romanian territory fills in the knowledge about spatial distribution of the genus *Hirudo* at international level (Utevsky *et al.*, 2010).

In the research area the distribution and dynamic of numerical density and biomass presented differences in time and space. The great number of leeches sampled in Chriloaia lake in May can indicate a high activity of leeches in search for their host. The presence of a rich fauna in the park area (mammals, amphibians) and the fact that Fundu Mare and Popa are strictly protected areas can offer the population of leeches the optimum necessary for their development. This hypothesis however is not validated by the small number of sampled individuals in the lakes Bordeiele and Gasca. Being lakes situated in the strictly protected area of the park, the numerical densities and biomass have registered very different values. Therefore in July in Gasca the number of sampled leeches has been 14 times bigger than the number sampled in Curcubeu (interconnected lakes through channels). The lakes Curcubeu and Vulpasu (disconnected lakes at sampling time) have registered very similar values (Fig. 4).

In the research area it has also been planned to identify the laid cocoons but unfortunately the search on the lake shore did not give any results. Nevertheless, the appearance of individuals with low biomass (0.02 g), in June (Chiriloaia) suggests changes in the structure of the population determined by the introduction of new individuals through newly-hatched leeches.

At national level, this type of study appears to be the first research dedicated to this category of aquatic invertebrates. Continuing research on-site could determine the main command factors and their ways of exercising pressure, which influence the dynamics of the *H. verbana* population.

References

Codell, C. K., 2001. Leech craft in nineteenth British medicine, *Journal of the Royal Society of medicine*, volume **94**.

Demirsoy, A., Kasparek, M., Akbulut, A., Durmuş, Y., 2001. Phenology of the medicinal leech, *Hirudo medicinalis* L., in north-western Turkey. *Hydrobiologia* **462**: 19–24.

Elliott, J. M. and Tullett, P. A., 1986. The effects of temperature, atmospheric pressure and season on the swimming activity of the medicinal leech*, Hirudo medicinalis* (Hirudinea: Hirudinidae), in a Lake District tarn. *Freshwater Biology,* **16**, 405.

Fira, V. and Nastasescu, M., 1977. *Zoologia nevertebratelor,* Editura Didactica si Pedagogica, Bucuresti.

Radu, V. G. and Radu, V. V., 1972. *Zoologia nevertebratelor* [pentru uzul studentilor]/Editura Didactica si Pedagogica, Bucuresti.

Sawyer, R. T., 1981. Why we need to save the medicinal leech. *Oryx* **16**, 165–168.

Scriban, I. A., 1904. *Contribution à la Faune des Hirudinées d'eau douce de Roumanie*, Editura Dacia, Jassy.

Scriban, I. A., 1910. *Contribution à l'anatomie et à l'histologie des hirudinées*, Editura Imprimerie Nationale, Jassy.

Siddall, M. E., Trontelj, P., Utevsky, S. Y., Nkamany, M., Macdonald, K. S. III, 2007. Diverse molecular data demonstrate that commercially available medicinal leeches are not *Hirudo medicinalis*. *Proc. R. Soc.* Lond. Ser. B. **274**: 1481–1487.

Trontelj, P., Sotler, M., Verovnik, R., 2004. Genetic differentiation between two species of the medicinal leech, *Hirudo medicinalis* the neglected *H. verbana*, based on random amplified polymorphic DNA. *Parasitol Res.* **94**: 118–124.

Trontelj, P., Utevsky, S. Y., 2005. Celebrity with a neglected taxonomy: molecular systematics of the medicinal leech (genus *Hirudo*). *Mol. Phylogenet. Evol*. **34**: 616–624.

Utevsky, S. Y., Trontelj, P., 2005. A new species of the medicinal leech (Oligochaeta, Hirudinida, Hirudo) from Transcaucasia and an identification key for the genus Hirudo. *Parasitol Res*. **98**: 61–66.

Utevsky, S., Zagmajster, M., Atemasov A., Zinenko O., Utevska O., Utevsky A. and Trontelj P., 2010. Distribution and status of medicinal leeches (genus *Hirudo*) in the Western Palaearctic: anthropogenic, ecological, or historical effects. *Aquatic Conserv: Mar. Freshw. Ecosyst*. **20**: 198–210.

Vadineanu, A., 2001. Lower Danube Wetlands System (LDWS), *Observatorio Medioambiental*, número **4**, 373-402.

Vadineanu, A., Adamescu, M., Cazacu, C., Bodescu, F. & Danielescu, S., 2001. Lower Danube Wetland System – Past and Future Management. *Earth Voice Romania*, Bucharest Univ., Dept. of Systems Ecology, Bucharest, pp. 74.

Whitaker, I. S., Rao, J., Izadi, D., Butler, P. E., 2004. Historical Article: *Hirudo medicinalis*: ancient origins of, and trends in the use of medicinal leeches throughout history. *British Journal of Oral and Maxillofacial Surgery* **42:** 133—137.

DIET OF GREAT CORMORANTS (*PHALACROCORAX CARBO SINENSIS*) IN INNER DANUBE DELTA, CHIRILOAIA COLONY

RELU CONSTANTIN GIUCA
Department of Systems Ecology and Sustainability, University of Bucharest, Splaiul Independenței 91-95, 050095, Bucharest, Romania, e-mail: giucarelu@yahoo.com

Abstract. The great cormorant (*Phalacrocorax carbo*) is a colonial, semi-migratory water bird that feeds almost entirely on fish and that is found in both inland and coastal waters. In the last decades its populations have been increasing throughout Europe, as a result of the legal protection measures and increased fish productivity due to eutrophication of aquatic ecosystems. The food requirements of these larger cormorant populations led to conflicts between bird protectionists and fisheries interests and therefore, the need for research on their diet and impact on fish populations.
Even though the diet and foraging behavior of cormorants has been studied before in different areas, there is no data concerning the interactions between cormorants and the fish communities in Inner Danube Delta (IDD). The diet of cormorants from Chiriloaia colony within IDD was studied using regurgitates analyses. The food samples were collected at the colony in May-July 2011 right after the morning feeding. The results show that low commercial value fish species within Cyprinids family represent the bulk of cormorants' diet, followed by Esocids.

Key words: great cormorant, *Phalacrocorax carbo*, diet, regurgitate, fish, Inner Danube Delta, piscivorous birds

Rezumat. Spectrul trofic al cormoranului mare (*Phalacrocorax carbo sinensis*) din Delta Interioară a Dunării, Colonia Chiriloaia. Cormoranul mare (*Phalacrocorax carbo*) este o pasăre colonială, semi-migratoare, ce se hrăneşte aproape exclusiv cu peşte, întalnită atât în apele continentale, cât şi în zonele de coastă. În ultimele decenii, populaţiile acestei specii au cunoscut o creştere continuă ca urmare a măsurilor de protecţie şi a creşterii productivităţii piscicole datorită eutrofizării ecosistemelor acvatice. Ca urmare a creşterii necesarului de hrană, au apărut conflicte de interese între protecţia păsărilor şi protecţia resursei piscicole, fapt ce a dus la necesitatea unor studii cu privire la spectrul trofic şi impactul asupra populaţiilor de peşti.
Deşi comportamentul de hrănire şi spectrul trofic au fost studiate în alte regiuni, totuşi nu există date despre interacţiunile dintre cormorani şi comunităţile piscicole din Delta Interioară a Dunării. Spectrul trofic al cormoranilor prezenţi în colonia Chiriloaia din Delta Interioară a Dunării a fost studiat, în perioada de cuibărire mai-iulie 2011, pe baza analizei prăzii regurgitate de păsări după hrănirea de dimineaţă. S-a constatat că speciile de peşte din familia Cyprinidae, cu valoare comercială scăzută, constituie principala sursă de hrană, fiind urmate de cele din familia Esocidae.

Cuvinte cheie: cormoranul mare, *Phalacrocorax carbo*, spectru trofic, regurgitat, peşte, Delta Interioară a Dunării, păsări ihtiofage

Introduction

The Great Cormorant (*Phalacrocorax carbo*) is a colonial, semi-migratory piscivorous bird found in both inland and coastal waters around the globe. In the last decades, its populations have been increasing throughout Europe, as a result of the legal protection measures and increased fish productivity due to eutrophication of aquatic ecosystems, leading to conflicts with fisheries and angling interests in some countries (Van Eerden *et al.*, 1995; Suter, 1995; Russell *et al.*, 1996; Cowx, 2003). The increasing trend was observed for Romanian populations too, which are represented by *Phalacrocorax carbo sinensis* subspecies, having the highest

concentration in the Danube Delta and Razim-Sinoe lagoon system (Năvodaru *et al.*, 2004).

Even though the diet and foraging behavior of cormorants has been studied before in different places, there is no data concerning the interactions between cormorants and the fish communities in Inner Danube Delta (IDD).

Materials and methods

Study area. Field work was conducted in Small Island of Braila (Fig. 1), a Long Term Socio Ecological Research site designated as a Wetland of International Importance in accordance with the Ramsar Convention, at a breeding colony located on the Chiriloaia Lake (45°11'58" N, 27°58'19" E) where the Great Cormorants are nesting in willow trees (*Salix* spp.) together with Pygmy Cormorants (*Phalacrocorax pygmeus*), Night Herons (*Nycticorax nycticorax*), Squacco Herons (*Ardeola ralloides*), Little Egrets (*Egretta garzetta*), Grey Herons (*Ardea cinerea*), Great Egrets (*Ardea alba*), Spoonbills (*Platalea leucorodia*) and Glossy Ibises (*Plegadis falcinellus*).

Diet of Great Cormorants was studied during the breeding season using regurgitates analyses, an easy and non-invasive method for collecting samples. The adults and the nestlings regurgitate their stomach content when disturbed during the visits to the colony, and if regurgitates are collected soon after the returning of the parents from the fishing areas, completely undigested fish can be found (Van Dobben, 1952; Linn & Campbell, 1992). The samples were collected at the colony under their nests in May - July 2011, after the morning feeding (10.00 - 12.00), stored in plastic bags and transported in a cool box to the lab for further analysis.

Fish preys were identified, intact specimens were measured to the nearest 1 cm and partly digested ones, that usually have an undamaged body but no head, were estimated in comparison with the intact ones.

Figure 1. Small Island of Braila.

Results and discussions

This is the first study on cormorant predation on a fish community in Inner Danube Delta. During the study period from May to July 2011, a total of 179 individual prey items were identified and measured, belonging to 7 different species and 5 families. The size range of the fish found was considerable, from 5 to 36 cm (Table 1).

Table 1. Fish species identified in the diet of *Phalacrocorax carbo sinensis.*

			Fish total length (cm)			
Scientific name	**Name**	**Family**	**Mean**	**Min**	**Max**	**N**
Carassius gibelio	Prussian carp	Cyprinidae	14	5	30	142
Abramis brama	Common bream	Cyprinidae	17	17	17	2
Scardinius erythrophtalmus	Rudd	Cyprinidae	17	12	22	4
Esox lucius	Pike	Esocidae	31	17	34	22
Perca fluviatilis	Perch	Percidae	16	10	20	5
Silurus glanis	Catfish	Siluridae	31	25	36	2
Lepomis gibbosus	Pumpkinseed	Centrarchidae	11	9	12	2

Cyprinids were dominant with 3 species, representing ~83% of all individual fish; within this family, Prussian carp (*Carassius gibelio*) was the most common prey species with individuals range from 5 to 30 cm long. Esocids (*Esox lucius*) were also well represented in the diet of cormorants with ~12% of fish prey, having lengths ranging from 17 to 34 cm. Other species (*Perca fluviatilis, Silurus glanis and Lepomis gibbosus*) were present in small number in the food of cormorants (Fig. 2).

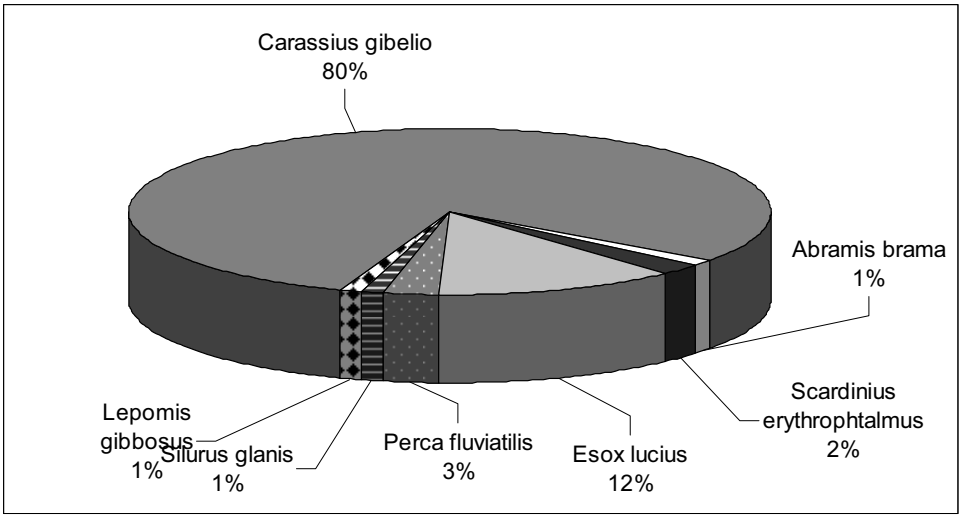


Figure 2. Percent occurrence of fish species identified in the diet of *Phalacrocorax carbo sinensis.*

The dominant size class for Prussian carp was found between 10-20 cm (Fig. 3) and for Pike 30-40 cm (Fig. 4).

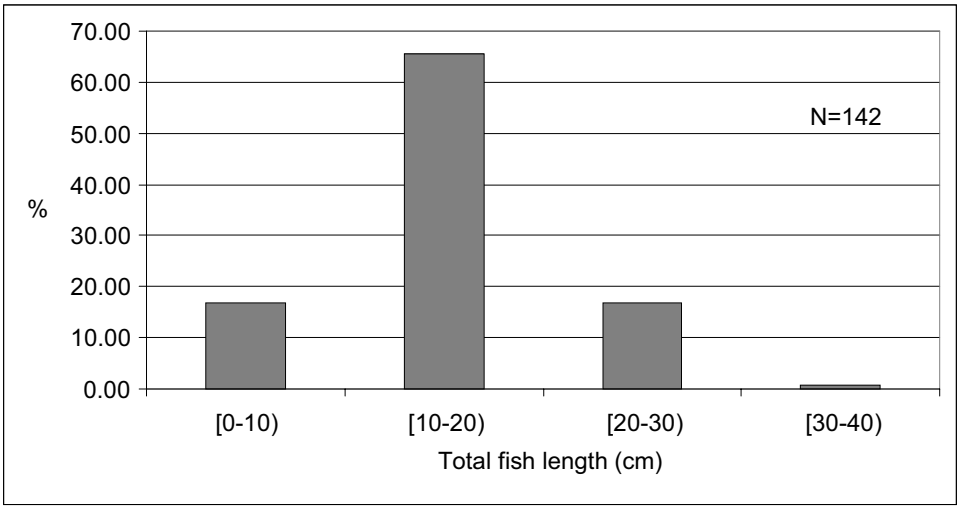


Figure 3. Size frequency of *Carassius gibelio* identified in the diet of *Phalacrocorax carbo sinensis.*

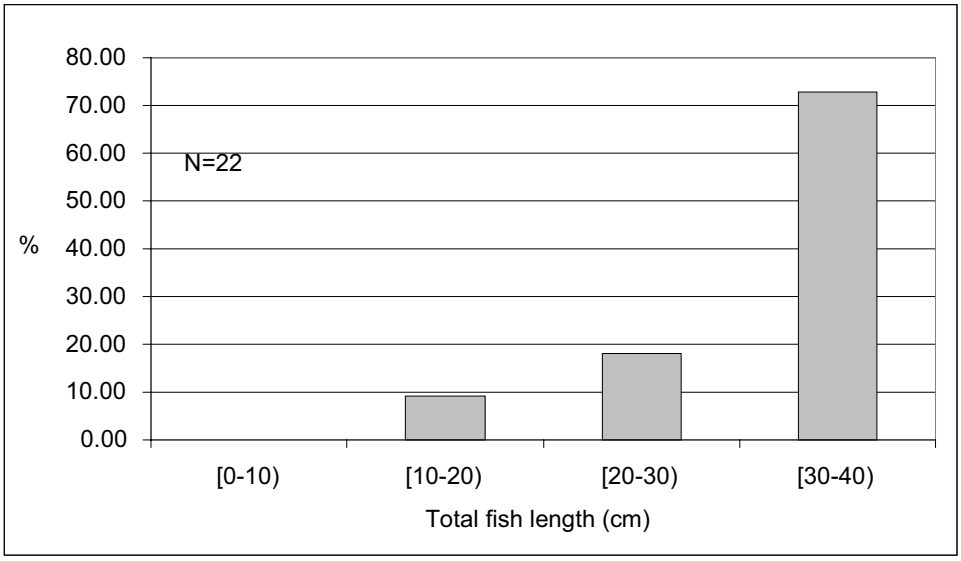


Figure 4. Size frequency of *Esox lucius* identified in the diet of *Phalacrocorax carbo sinensis.*

The study revealed that cormorants can also forage on large individuals of *Esox lucius* and *Silurus glanis*, with lengths of 34 and 36 cm respectively.

Low commercial value fish species are dominant in the diet of *Phalacrocorax carbo sinensis* (Fig. 5).

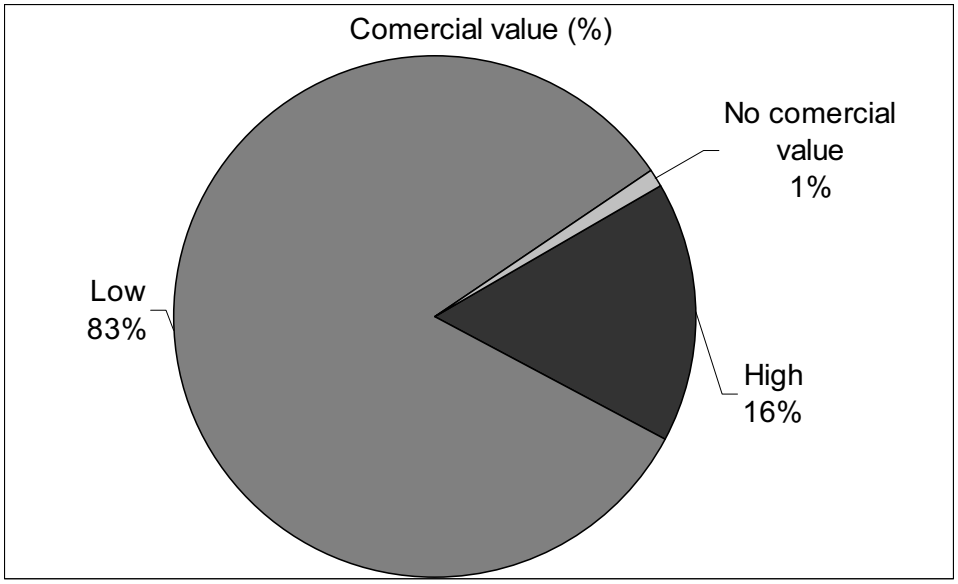


Figure 5. Commercial value of fish identified in the diet of *Phalacrocorax carbo sinensis.*

Conclusions

Great Cormorant diet includes different fish species, but only a few of them usually compose the bulk of their diet. The prevalence of few fish species in their diet has also been observed in other countries (Boldreghini *et al.*, 1997; Lorentsen *et al.*, 2004; Liordos & Goutner, 2007), being characteristic to the opportunistic foraging behavior of this piscivorous bird (Johnsgard, 1993). The preys that are most available and abundant in an area and season will be found predominantly in their diet (Grémillet *et al.*, 2001) since Great Cormorants are hunting prey with very high densities. Thus, the link between foraging performance and fish abundance is stronger than in any other diving bird species (Grémillet & Wilson, 1999).

Fish of low commercial value and of small to medium sizes mainly comprises the diet of *Phalacrocorax carbo sinensis*, as indicated in other European studies (Keller, 1995; Veldkamp, 1995; Mellin & Krupa, 1997; Carpentier *et al.*, 2003).

Economic impact may occur in small areas of high fish concentrations such as fish farms, wintering reservoirs, small lakes and rivers (Kirby *et al.*, 1996; Leopold *et al.*, 1998; Wright, 2003).

Acknowledgments

This work was supported by project: POSDRU/88/1.5/S/61150 “Doctoral Studies in the field of life and earth sciences”, project co-financed through Sectorial Operational Program for the Development of Human Resources 2007-2013 from European Social Fund.

References

Boldreghini, P., Santolini, R., Pandolfi, M., 1997. Abundance and frequency of occurrence of prey-fish in the diet of cormorants *Phalacrocorax carbo* in the Po Delta (Northern Italy) during the wintering period. *Ekologia Polska*, **45**: 191-196.

Carpentier, A., Paillisson, J.M., Marion, L., 2003. Assessing the interaction between cormorants and fisheries: the importance of fish community change. Pages 187-195 in *Interactions Between Fish and Birds: Implications for Management* (I. G. Cowx, Ed.). Fishing News Books, Blackwell Science, Ltd., Oxford.

Cowx, I. G., 2003. *Interactions between fish and birds: Implications for management.* Fishing News Books, Blackwell Science Ltd., Oxford.

Grémillet, D., Wanless, S., Carss, D.N., Linton, D., Harris, M.P., Speakman, J.R., Le Maho, Y., 2001. Foraging energetics of arctic cormorants and the evolution of diving birds. *Ecology Letters*, **4**: 180-184.

Grémillet, D., Wilson, R.P., 1999. A life in the fast lane: energetics and foraging strategies of the Great Cormorant. *Behavioural Ecology*, **10**: 516-524.

Johnsgard, P.A., 1993. *Cormorants, Darters, and Pelicans of the World*. Smithsonian Institution Press, Washington.

Keller, T., 1995. Food of cormorants *Phalacrocorax carbo sinensis* wintering in Bavaria, southern Germany. *Ardea*, **83**: 185-192.

Kirby, J. S., Holmes, J.S., Sellers, R.M., 1996. Cormorants *Phalacrocorax carbo* as fish predators: an appraisal of their conservation and management in Great Britain. *Biological Conservation*, **75**: 191-199.

Leopold, M. F., Van Damme, C.J.G., Van der Veer, H.W., 1998. Diet of cormorants and the impact of cormorant predation on juvenile flatfish in the Dutch Wadden Sea. *Journal of Sea Research*, **40**: 93-107.

Linn, I. J., Campbell, K. L. I., 1992. Interactions between white-breasted cormorants *Phalacrocorax carbo* (Aves: Phalacrocoracidae) and the fisheries of Lake Malawi. *Journal of Applied Ecology*, **29**: 619-634.

Liordos, V., Goutner, V., 2007. Spatial patterns of winter diet of the Great Cormorant in coastal wetlands of Greece. *Waterbirds*, **30**: 103-111.

Lorentsen, S.H., Grémillet, D., Nyomen, G.H., 2004. Annual variation in diet of breeding Great Cormorants: does it reflect varying recruitment of gadoids? *Waterbirds*, **27**: 161-169.

Mellin, M., Krupa, R., 1997. Diet of Cormorant, based on the analysis of pellets from breeding colonies in NE Poland. *Proceedings IV European Conference on Cormorants, Bologna, Italy. Supplemento alle Richerche di Biologia della Selvaggina*, **XXVI**: 511-516.

Năvodaru, I., Kiss, J. B., Cernişencu, I., 2004. Fishery and piscivorous birds forced to sustain together in Danube Delta, Romania (review). *Studii şi cercetări ştiinţifice. Biologie, Serie nouă*. Universitatea din Bacău, **8**: 128-139.

Russell, I. C., Dare, P. J., Eaton, D.R., Armstrong, J.D., 1996. *Assessment of the problem of fish-eating birds in inland fisheries in England and Wales.* Report to the Ministry of Agriculture, Fisheries and Food, project number VC0104. MAFF, London.

Suter, W., 1995. The effect of predation by wintering Cormorants *Phalacrocorax carbo* on Grayling *Thymallus thymallus* and Trout (Salmonidae) populations: two case studies from Swiss rivers. *Journal of Applied Ecology*, **32**: 29-46.

Van Dobben, W. H., 1952. The food of the Connorant in The Netherlands. *Ardea*, **40**: 1-63.

Van Eerden, M. R., Koffijberg, K., Platteeuw, M., 1995. Riding on the crest of the wave: possibilities and limitations for a thriving population of migratory Cormorants *Phalacrocorax carbo* in man-dominated wetlands. *Ardea*, **83**: 1-9.

Veldkamp, R., 1995. Diet of Cormorants *Phalacrocorax carbo sinensis* at Wanneperveen, The Netherlands, with special reference to Bream *Abramis brama*. *Ardea*, **83**: 143-155.

Wright, G. A., 2003. Impact of cormorants on the Loch Leven trout fishery and the effectiveness of shooting as mitigation. Pages 288-297 in *Interactions Between Fish and Birds: Implications for Management* (I. G. Cowx, Ed.). Fishing News Books, Blackwell Science, Ltd., Oxford.

FLORA AND VEGETATION DIVERSITY IN THE STRICTLY PROTECTED AREA ROSCA - BUHAIOVA (DANUBE DELTA)

NECULAI STEFAN[1], ION SARBU[1] and JENICA HANGANU[2]
[1] "Alexandru Ioan Cuza" University of Iasi, Bulevardul Carol I, Nr. 20A, Iasi, Romania;
[2] Danube Delta National Institute, Strada Babadag, Nr. 165, Tulcea, Romania, e-mail: hanganu@indd.tim.ro

Abstract. In the north-eastern part of the fluvial Danube Delta lies the strictly protected area Roşca-Buhaiova where 28 associations and one subassociation of aquatic and paludal vegetation have been identified. These cenotaxa belong to *Lemnetea*, *Potamogetonetea pectinati* and *Phragmitetea* classes.

Key words: Danube Delta, reserve, vegetation, aquatic, paludal

Rezumat. Diversitatea florei şi vegetaţiei în zona strict protejată Roşca-Buhaiova (Delta Dunării). În partea de nord-est a Deltei Dunării fluviale se află zona strict protejată Roşca-Buhaiova unde au fost identificate 28 asociaţii şi o subasociaţie a vegetaţiei acvatice şi palustre. Aceşti cenotaxoni aparţin claselor *Lemnetea*, *Potamogetonetea pectinati* şi *Phragmitetea*.

Cuvinte cheie: Delta Dunării, rezervaţie, vegetaţie, acvatic, paludal

Introduction

The Roşca-Buhaiova lake complex is located in the Matiţa Depression, in the north-eastern part of the Danube Delta. It has an area of approximately 9625 ha, starting from the Chilia branch (in the North), between the Chilia sand bank (in the West) and the Letea sand bank (in the East), being limited in the South by Dunărea Veche (the big "M"). It has been present on the Romanian natural reserves list as early as the 1970's, and starting from 1979 it is a Biosphere Reserve. It includes a large diversity of representatives habitats for the fluvial delta (floating and fixed reed islands, marshes, lakes, sand banks, natural brooks, floodplains, groves of willow trees, etc.). The centre of this reserve with a full protection regime consists of Roşca and Buhaiova lakes. The elevations close to 0 m are dominant, this being the reason for which approximately 90% of this area is permanently submerged. Because the communication with Chilia and Sulina branches is difficult and the flooding water brings few mineral suspensions, an extremely intense process of peat formation takes place. Organic soils are dominant in the area, next to which limnosols, gley soils (marshy soils, submerged soils or peaty soils) can also be found; alluvial deposits only appear on small areas, in Lopatna brook's nearby surroundings, in the northern part of this protected area.

The flora exhibits a relatively reduced diversity, the floristic inventory consisting of 92 aquatic and paludal species out of which two are included in the Habitats Directive 92/43 (*Salvinia natans* and *Trapa natans*).

The vegetation developed especially on the peat deposits, the *Scirpo-Phragmitetum* W. Koch 1926 association being dominant, with *Salix cinerea* and *Thelypteris palustris*, this area being 60-95% covered by a diverse aquatic vegetation (Ştefan *et al*., 1995).

The phytocenological conspectus consists of 29 coenotaxa (28 associations and one subassociation (Hanganu *et al*.; Ştefan & Sârbu, 2002) which belong to three classes of vegetation (*Lemnetea* O. de Bolós et Masclans 1955, *Potamogetonetea pectinati* Tx et Prsg. 1942, *Phragmitetea* Tx. et Prsg. 1942) (Coldea, 1997; Sanda & Popescu, 1973):

LEMNETEA O. de Bolós et Masclans 1955
LEMNETALIA MINORIS O. de Bolós et Masclans 1955
Lemnion minoris O. de Bolós et Masclans 1955
1. *Lemnetum minoris* Soó 1927
2. *Lemnetum trisulcae* Knapp et Stoffer 1962
3. *Lemno minoris – Spirodeletum* W. Koch 1954
Lemno - Salvinietum natantis Slavnic 1956
4. *Lemno – Salvinietum natantis* Mijavaki et J.Tx.1960
5. *Lemno – Azolletum carolinianae* Nedelcu 1967
LEMNO – UTRICULARIETALIA Pass. 1978
Utricularion vulgaris Pass. 1964
6. *Lemno – Utricularietum vulgaris* Soó 1928
HYDROCHARIETALIA Rübel 1933
Hydrocharition Rübel 1933
7. *Lemno – Hydrocharietum morsus-ranae* (Oberd. 1957)Pass. 1978
8. *Stratiotetum aloides* Nowinnski 1030
Ceratophyllion demersi Soó 1927
9. *Ceratophylletum demersi* Hild. 1956

POTAMOGETONETEA PECTINATI R. Tx. et Prsg. 1942
POTAMOGETONETALIA PECTINATI W. Koch 1926
Potamogetion lucentis Rivas Martinez 1973
10. *Elodeetum nuttalii* Ciocârlan *et al.* 1997
11. *Potamogetonetum lucentis* Hueck 1931
12. *Potamogetonetum perfoliati* Koch 1926 em. Pass. 1964
Potamogetion pussili Vollmar em Hejný 1978
13. *Potamogetonetum crispi* Soó 1927
14. *Potamogetonetum pectinati* Carstensen 1955
15. *Potamogetonetum trichoides* J. Et R. Tx. 1955
Nymphaeion albae Oberd. 1957
16. *Nympaeetum albo-candidae* Pass. 1957
17. *Polygonetum amphibii* Soó 1927
18. *Potamogetonetum natantis* Soó 1927
19. *Nymphoidetum peltatae* (All. 1922) Bellot 1951
20. *Trapo – Nymphoidetum* Oberd. 1957
21. *Trapetum natantis* V. Kárpati 1963
22. *Myriophyllo verticillati – Nupharetum luteae* W. Koch 1926

PHRAGMITETEA Tx. et Prsg. 1942
PHRAGMITETALIA W. Koch 1926 em. Pign. 1953

P h r a g m i t i o n c o m m u n i s W. Koch 1926
23. *Scirpo – Phragmitetum* W. Koch 1926
- subass. *salicetosum cinereae* Ştefan et al. 1995
24. *Thelypterido – Phragmitetum* Kuiper 1958
25. *Typhetum angustifoliae* Pign. 1953
26. *Glycerietum maximae* Hueck 1931
P h a l a r i d i o n a r u n d i n a c e a e Kopecký 1961
27. *Phalaridetum arundinaceae* (W. Koch 1926) Libbert 1931
OENANTHETALIA AQUATICAE Hejný in Kopecký ex Hejný1965
28. *Oenantho – Rorippetum* Lohmeyer 1950

In the areas where the water surface of the lakes is interrupted by big reed islands it can be noticed the dominance of emerged phytocoenoses, which have a preference for stagnant or slow running waters from *Lemnion minoris* O. de Bolós et Masclans 1955 (Table 1., as. 1-2) and *Lemno-Salvinion natantis* Slavnic 1956 (Table 1., as. 3-5) alliances. In the places where the water is deeper and has a larger quantity of degraded organic matter, phytocoenoses from *Utricularion vulgaris* Passarge 1978 alliance (Table 1., as. 6) occur and also unfixed and emerged associations from the *Hydrocharition* Rübel 1933 alliance (Table 1., as. 7-8) or submerged associations from the *Ceratophyllion demersi* Soó 1927 alliance (Table 1., as. 9).

In open and still sites, aquatic emerged or submerged phytocenoses from the *Potamogetonetalia pectinati* W. Koch 1926 order (Table 2., as. 1-6) occur and near the banks or at the edges of the reed beds there are relatively large areas occupied by rooted phytocenoses with emerged or submerged leaves from the *Nymphaeion albae* Oberd. 1957 alliance (Table 3., as. 1-7).

On the floodplains, at the edges of the marshes or in small depressions with stagnant water, on alluvial soils or alluvial protosols, rich in organic matter that are slightly acidic up to neutral or slightly alkaline, numerous hygrophyte associations vegetate, with few species, from the *Phragmitetea* Tx. et Prsg. 1942 class (Table 4., as. 1-6).

Table 1. *Lemnetea* O. de Bolós et Masclans 1955.

Association	1			2		3			4			5		6		7		8			9	
Vegetation cover (%)	85	75	90	70	65	90	90	85	85	90	85	80	90	75	70	80	75	65	70	65	75	80
Water depth (cm)	120	130	80	60	70	100	90	120	100	120	90	40	60	150	140	90	110	180	150	160	130	150
Sample area (m^2)	10	25	8	4	4	10	15	25	10	10	25	4	6	10	15	8	10	4	4	6	10	15
Lemnion																						
Lemna minor	5	4	5	1	1	3	2	3	2	1	1	2	1	1	+	2	1	+	+	+	+	+
Lemna trisulca	-	-	-	4	3	-	+	-	-	-	-	-	-	-	+	-	-	-	-	-	-	-
Lemna gibba	-	+	-	-	+	-	-	-	-	-	+	-	-	-	-	-	-	-	-	-	-	-
Wolffia arrhiza	-	-	-	-	-	-	+	-	-	-	-	+	-	-	-	-	-	-	-	-	-	-
Hydrocharition																						
Salvinia natans	-	+	+	-	+	+	-	-	3	4	4	+	-	+	-	1	1	+	+	-	+	+
Stratiotes aloides	-	-	-	+	-	-	+	-	+	-	-	-	-	+	-	+	-	3	4	3	-	-
Hydrocharis morsus ranae	-	-	-	-	-	-	-	+	-	-	-	-	+	-	-	3	3	-	-	-	-	-
Ceratophyllion demersi																						
Ceratophyllium demersum	+	-	-	-	+	-	-	+	-	+	-	-	-	+	1	-	-	-	-	1	4	4
Lemnetea																						
Azolla filiculoides	-	-	-	-	+	-	-	-	-	-	-	3	4	-	-	-	-	-	-	-	-	-
Utricularia vulgaris	-	-	+	-	-	-	-	-	-	-	-	-	-	4	4	-	-	-	+	-	+	+
Spirodela polyrhiza	+	+	+	-	+	3	3	3	+	-	+	1	+	-	+	-	+	-	+	+	-	+
Potamogetonetea pectinati																						
Elodea nuttalii	-	-	+	-	+	-	+	+	-	-	-	+	-	+	+	-	-	+	-	-	+	1
Elodea Canadensis	-	-	-	+	-	-	-	-	-	+	+	-	-	-	-	-	-	-	-	-	-	-
Myriophyllum verticillatum	-	-	-	-	-	-	-	-	+	-	-	-	+	-	-	+	-	-	-	-	-	-

Ranunculus trichophyllus	-	-	-	+	-	+	1	-	-	-	-	+	-	-	-	-	-	-	-	-	-	-
Potamogeton natans	+	-	-	-	-	-	-	-	-	-	+	-	-	+	-	+	+	-	-	-	-	-
Potamogeton pectinatus	-	+	-	-	-	-	-	+	-	-	-	-	-	+	+	-	-	+	+	-	+	-
Potamogeton trichoides	-	-	-	+	1	-	-	1	+	+	+	-	-	-	-	-	-	+	+	1	-	-
Potamogeton crispus	-	-	-	-	-	+	-	-	-	-	-	+	-	+	+	-	-	+	-	1	-	+
Trapa natans	+	-	-	-	-	+	1	+	1	-	-	-	-	-	-	+	1	1	-	-	-	-
Polygonum amphibium	-	-	-	-	-	-	-	-	-	+	-	+	+	-	-	-	-	-	-	-	+	-
Nymphaea alba	-	-	+	-	-	-	-	1	-	-	-	1	-	-	-	1	+	1	-	+	-	-
Nuphar lutea	-	-	-	-	-	+	-	-	-	-	-	-	-	+	+	-	-	-	-	-	+	-
Nymphoides peltata	-	-	-	-	-	-	+	-	-	-	-	-	+	-	-	-	-	-	-	-	-	-
Phragmitetea																						
Typha angustifolia	-	-	-	-	-	-	-	-	-	-	-	+	-	-	-	+	-	-	-	-	-	-
Phragmites australis	-	+	-	-	+	-	+	-	-	+	-	+	+	-	-	-	-	-	-	-	-	-
Oenanthe aquatic	-	-	-	+	-	-	-	-	-	-	-	+	-	-	-	+	-	-	-	-	-	-
Alisma plantago-aquatica	-	-	-	-	+	-	-	-	-	-	-	+	+	-	-	-	-	-	-	-	-	-
Mentha aquatic	-	-	-	-	-	-	-	-	-	-	-	-	+	-	-	-	-	-	-	-	-	-
Glyceria maxima	-	-	-	+	-	-	-	-	-	-	-	+	-	-	-	-	-	-	-	-	-	-
Berula erecta	-	-	-	-	+	-	-	-	-	-	-	+	+	-	-	-	-	-	-	-	-	-

Associations : **1** = *Lemnetum minoris* Soó 1927; **2** = *Lemnetum trisulcae* Knapp et Stoffers 1962; **3** = *Lemno-Spirodeletum* W. Koch 1954; **4** = *Lemno-Salvinietum* Mijavaki et J.Tx. 1962; **5** = *Lemno-Azolletum* (van Langedock 1935) Br.-Bl.1952; **6** = *Lemno-Utricularietum vulgaris* Soó 1928; 7 = *Lemno-Hydrocharietum* (Oberd. 1957)Pass.1978; **8** = *Stratiotrtum aloides* Nowinski 1930; **9** = *Ceratophylletum demersi* Hild. 1956

Table 2. *Potamogetonetea pectinati* Tx. et Prsg. 1942.

Association	1			2		3			4		5				6		
Vegetation cover (%)	75	85	90	60	55	65	70	70	65	60	90	85	90	95	80	65	85
Water depth (cm)	60	70	70	100	90	120	110	120	140	125	60	55	80	100	110	80	90
Sample area (m^2)	10	6	8	4	10	10	6	6	4	8	12	15	20	20	8	10	10
Potamogetion lucentis																	
Elodea nuttalii	4	4	4	-	-	+	-	-	+	-	-	-	-	-	-	-	+
Elodea Canadensis	+	1	-	+	-	+	-	-	-	+	-	-	-	-	-	-	-
Potamogeton lucens	-	-	+	4	3	-	-	1	-	-	-	-	-	-	+	-	-
Potamogeton crispus	-	+	-	-	-	-	+	-	4	3	-	+	-	+	-	-	-
Potamogeton perfoliatus	-	-	-	-	-	3	4	3	-	-	-	-	+	-	-	-	-
Myriophyllum verticillatum	+	-	-	-	-	-	-	+	-	-	-	-	-	-	-	1	-
Potamogetonetalia pectinati																	
Potamogeton pectinatus	+	+	-	+	-	1	-	1	+	1	5	4	4	5	-	1	+
Potamogeton trichoides	+	-	1	+	+	+	+	-	+	1	-	1	+	-	4	3	5
Potamogeton natans	-	-	-	-	+	-	-	-	-	-	-	-	-	-	-	-	-
Ranunculus trichophyllus	-	-	-	-	-	+	-	-	-	-	+	-	-	-	-	-	-
Nymphaeion albae																	
Nymphaea alba	-	+	-	-	-	-	-	-	+	-	-	-	-	+	-	1	-
Nymphaea candida	-	-	-	-	-	+	-	-	-	-	-	-	+	-	-	-	-
Nuphar lutea	-	-	-	-	-	-	+	-	-	+	-	-	-	-	-	-	-
Polygonum amphibium f. aquaticum	+	-	-	-	-	-	+	+	-	-	+	+	-	-	+	-	-
Trapa natans	-	+	1	-	-	-	-	-	+	-	-	-	-	-	+	-	-
Lemnetea																	
Lemna minor	+	+	+	-	-	+	-	+	-	1	+	+	+	-	+	+	+
Lemna trisulca	-	-	-	-	+	-	-	-	-	+	+	-	-	-	-	-	-
Spirodela polyrrhyza	+	+	-	+	-	-	+	+	+	+	-	+	+	+	-	-	+
Salvinia natans	-	+	-	-	+	+	-	+	-	-	+	+	+	+	-	-	-
Stratiotea aloides	-	-	-	-	-	-	-	-	-	+	-	+	-	-	-	-	-
Utricularia vulgaris	-	-	+	-	-	-	-	-	+	-	-	-	1	+	-	1	-
Phragmitetea australis																	
Phragmitea australis	+	-	-	-	+	-	-	-	-	+	+	-	-	-	-	+	-
Typha angustifolia	-	-	-	-	-	-	-	-	-	-	+	-	-	-	-	-	-
Butomus umbellatus	-	-	+	-	-	-	-	-	-	-	-	-	-	-	-	+	-
Alisma plantago aquatic	-	-	-	-	-	-	-	-	-	-	+	+	-	-	-	-	-
Sagittaria sagittifolia	+	-	+	+	-	-	-	-	-	-	-	-	+	-	-	-	-

Glyceria maxima	-	-	-	-	-	-	-	-	-	-	-	+	-	-	-	+	-
Eleocharis palustris	-	-	-	-	-	-	-	-	-	-	+	-	-	-	-	-	-
Rorippa amphibian	-	-	-	-	-	-	-	-	-	-	-	+	-	-	-	+	-

Associations: **1** = *Elodeetum nuttalii* Ciocîrlan et al.1997; **2** = *Potamogetonetum lucentis* Hueck 1931; **3** = *Potamogetonetum perfoliati* W.Kock 1926 em.Pas. 1964; **4** = *Potamogetonetum crispi* Soó 1927; **5** = *Potamogetonetum pectinati* Carstensen 1955; **6** = *Potamogetonetum trichoides* J. et R. Tx. 1965

Table 3. *Nymphaeion albae* Oberd. 1957.

Association	1			2		3		4		5		6				7		
Vegetation cover (%)	80	85	80	70	70	60	70	85	80	90	85	95	95	90	95	75	85	75
Water depth (cm)	130	150	120	90	80	120	140	70	60	80	110	90	70	70	85	120	100	95
Sample area (m^2)	50	60	25	6	10	4	6	10	15	20	25	25	50	50	40	20	12	8
Nymphaeion albae																		
Nymphaea alba	3	4	3	1	-	1	1	+	-	-	-	+	-	+	-	-	-	-
Nymphaea candida	2	1	1	+	-	+	-	1	-	+	-	1	+	-	+	-	-	-
Nuphar lutea	-	-	-	-	+	-	-	-	1	+	1	-	+	-	-	3	4	3
Polygonum amphibium f. aquaticum	-	+	-	3	4	-	-	+	-	-	-	+	-	-	-	+	-	+
Potamogeton natans	-	-	1	-	-	3	4	-	-	3	2	4	5	4	4	-	-	-
Nymphoides peltata	-	-	-	-	+	-	-	4	4	1	2	+	-	-	-	-	-	-
Potamogetonetea pecrinati																		
Myriophyllum verticillatum	-	-	+	-	-	-	-	-	+	-	-	-	-	+	-	3	2	2
Elodea nuttalii	+	-	1	+	+	-	-	-	-	+	+	-	-	+	+	-	+	+
Elodea Canadensis	-	-	-	+	-	-	+	-	-	-	+	+	-	-	-	+	-	-
Ceratophyllum demersum	-	+	-	-	-	-	-	+	-	-	-	-	-	-	-	-	-	-
Potamogeton perfoliatus	-	-	-	+	-	-	-	-	-	-	-	-	+	-	-	-	-	-
Potamogeton trichoides	+	-	+	1	+	1	1	-	-	-	+	-	+	-	-	+	+	+
Potamogeton pectinatus	-	+	-	+	-	+	1	+	1	1	-	+	+	+	-	-	-	+
Ranunculus trichophyllus	-	-	-	+	-	+	-	-	-	+	-	-	-	-	-	-	-	-
Lemnetea																		
Lemna minor	+	+	+	1	1	+	+	+	1	1	1	1	+	2	2	+	+	1
Lemna trisulca	+	-	-	-	-	+	+	-	-	-	-	-	-	-	-	+	+	-
Spirodela polyrrhiza	-	+	+	+	-	+	-	+	+	+	+	-	-	-	+	-	-	-
Salvinia natans	-	+	-	-	+	+	+	-	+	1	1	+	1	-	1	+	+	2

Utricularia vulgaris	-	-	-	+	-	-	+	-	-	-	+	-	-	-	-	-	-	-
Azolla filiculoides	-	+	-	-	-	-	-	-	-	-	-	-	+	-	-	-	-	-
Phragmitetea australis																		
Typha angustifolia	-	-	-	+	-	-	-	+	-	-	-	+	-	+	-	-	-	-
Schoenoplectus lacustris	-	-	-	+	-	-	-	-	+	-	-	-	-	-	-	-	-	-
Glyceria maxima	-	-	-	+	-	-	-	+	+	-	-	-	-	-	-	-	-	-
Cicuta virosa	-	+	-	-	-	+	-	-	-	+	-	-	-	-	-	+	-	-
Phragmites australis	+	-	+	-	+	+	-	+	+	-	-	-	+	-	+	-	-	+
Rorippa amphibian	-	-	-	-	+	-	-	-	+	+	-	-	+	-	-	-	+	-
Stachys palustris	-	-	-	-	-	-	-	+	-	-	-	-	-	-	-	-	-	-
Alisma plantago-aquatica	-	-	-	-	-	-	-	+	+	-	-	-	+	-	-	-	-	-
Sparganium neglectum	-	-	+	-	-	-	-	-	+	-	-	-	-	+	-	-	-	-
Butomus umbellatus	-	-	-	-	+	-	-	-	-	-	-	+	-	-	+	-	-	-
Valisneria spiralis	-	-	-	-	-	-	+	-	-	-	-	-	-	-	-	-	-	-

Associations: **1** = *Nymphaeetum albo-candidae* Pass. 1957; **2** = *Polygonetum amphibii* Soó 1927; **3** = *Potamogetonetum natantis* Soó 1927; **4** = *Nymphoidetumpeltatae* (Allorge 1922) Bellot 1951; **5** = *Trapo – Nymphoidetum* Oberd. 1957: **6** = *Trapetum natantis* V.Kárpati 1963; **7** = *Myriophyllo verticillati – Nupharetum luteae* W.Koch 1926

Table 4. *Phragmitetea* Tx. er Prsg. 1942.

Association	1					2			3				4				5			6		
Vegetation cover (%)	95	100	1100	90	95	75	75	85	90	85	80	85	95	90	95	90	85	90	85	70	75	70
Sample area (m^2)	100	100	100	100	100	100	100	100	50	100	100	100	100	50	100	25	100	100	25	50	100	50
Phragmition et Phragmitelalia																						
Phragmites australis	4	5	5	4	5	4	3	4	4	4	4	4	+	+	1	+	1	+	1	+	1	+
Schoenoplectus lacustris	1	+	+	+	-	+	+	-	+	-	-	+	-		-	-	-	-	-	-	-	-
Oenanthe aquatic	+	-	-	+	-	-	+	-	+	-	-	-	-	-	+	-	-	-	-	3	3	2
Rorippa amphibian	-	+	-	+	-	+	-	+	-	+	+	-	-	+	-	-	-	-	-	2	2	3

Glyceria maxima	+	-	+	-	+	-	+	+	-	-	+	-	5	4	5	4	-	+	-	+	+	-
Ranunculus lingua	-	+	+	-	+	-	+	-	+	+	+	+	-	+	-	+	-	-	-	-	+	-
Senecio paludosus	+	-	-	+	-	+	+	+	-	+	+	+	+	-	-	+	-	-	+	+	-	+
Sium latifolium	-	-	+	-	-	+	+	-	+	-	+	+	-	+	+	-	-	-	-	-	+	-
Stachys palustris	+	+	+	-	+	+	-	+	+	+	+	-	+	+	-	+	+	-	+	+	+	+
Typha angustifolia	1	-	+	1	+	-	+	-	-	+	-	-	+	-	+	-	-	+	-	+	+	-
Typha latifolia	-	+	-	-	-	+	+	+	-	+	+	-	-	-	-	-	-	-	-	-	-	+
Cicuta virosa	-	-	+	-	-	-	+	+	-	+	+	+	-	+	-	-	-	-	-	1	+	+
Iris pseudacorus	-	-	+	+	-	-	+	-	+	-	-	+	+	-	+	+	-	-	-	+	-	-
Mentha aquatic	1	+	-	+	1	+	1	-	+	+	-	+	-	+	+	+	+	-	+	-	+	+
Magnocaricetalia elatae																						
Thelypteris palustris	-	-	-	-	-	+	-	-	2	1	2	1	-	-	-	-	-	-	-	-	-	-
Carex elata	-	+	+	-	+	-	-	-	+	+	-	+	-	-	+	-	-	-	-	+	-	-
Eleocharia palustris	+	-	+	-	-	-	-	-	-	-	-	-	+	+	-	+	-	-	-	-	-	+
Cirsium canum	-	-	-	-	-	-	-	-	-	-	-	-	-	-	+	-	+	-	-	-	-	-
Galium palustre	+	+	-	+	+	-	+	+	+	-	-	-	+	+	+	-	+	-	+	-	+	+
Lysimachia vulgaris	-	+	-	-	-	+	+	-	+	-	+	-	+	-	+	-	+	-	+	-	+	+
Lythrum salicaria	-	-	+	+	-	+	+	-	+	-	-	-	+	+	-	+	+	-	+	+	-	+
Phalaris arundinacea	-	-	-	-	-	-	+	-	-	+	-	-	+	-	-	+	4	5	4	-	-	-
Scutellaria galericulata	+	-	+	-	-	+	-	+	+	+	+	-	-	+	-	+	-	-	-	+	-	-
Carex riparia	+	-	-	1	-	+	2	1	+	-	+	+	-	+	-	+	+	-	-	+	-	+
Carex acutiformis	1	-	+	1	+	1	1	+	+	-	+	+	+	-	+	-	+	+	+	-	+	-
Phragmitetea																						
Lycopus europaeus	-	-	+	-	+	+	+	-	+	-	+	-	+	-	-	-	-	+	-	+	+	-

Bolboschoenus maritimus	-	-	-	+	-	-	+	-	-	-	-	-	-	+	-	+	+	+	+	-	-	+
Rumex hydrolapathum	-	+	-	-	+	+	-	+	-	-	+	-	+	+	-	-	-	-	+	+	+	
Solanum dulcamara	-	+	-	-	-	1	+	+	-	+	-	-	+	-	-	-	-	-	-	+	-	-
Alisma plantago/aquatic	-	-	-	+	-	-	+	-	+	+	-	+	-	+	+	+	-	+	-	-	+	-
Sparganium erectum	-	-	-	-	-	-	-	+	-	+	-	-	+	+	-	-	-	-	-	-	-	+
Berula erecta	+	-	-	+	+	-	-	+	-	-	+	-	+	-	-	-	-	+	-	-	-	-
Butomus umbellatus	-	-	-	-	-	+	-	+	-	+	+	+	-	+	-	-	-	-	-	+	+	-
Carex pseudocyperus	-	-	+	-	-	-	+	-	+	+	+	-	+	+	-	-	-	-	-	-	-	-
Veronica anagallis/aquatic	+	-	+	-	-	+	-	+	-	+	-	-	-	+	-	-	-	-	-	-	+	-
Symphytum officinale	+	+	1	-	+	-	+	+	-	+	+	-	-	+	+	+	-	-	+	-	+	+
Myosotis scorpioides	+	-	+	-	-	-	+	-	+	+	+	+	-	+	-	+	+	-	-	+	-	+
Aliae																						
Salix cinerea	-	+	-	-	+	1	2	1	-	+	-	+	-	-	+	-	-	+	-	+	-	-
Calystegia sepium	-	-	+	-	+	1	+	1	-		-	-	+	-	+	-	-	+	-	-	-	
Nymphaea alba	-	-	-	+	-	-	-	-	+	+	-	-	-	-	-	-	-	-	-	+	-	-
Nymphaea candida	-	+		-	-	-	-	-	-	-	+	-	-	+	-	-	-	-	-	-	-	+
Potamogeton pectinatus	-	+	-	-	-	-	-	+	-	-	-	-	-	-	-	-	-	-	-	-	+	-
Potamogeton natans	-	-	+	-	-	-	-	-	-	+	-	-	-	-	-	-	-	-	-	-	+	-
Polygonum amphibium	-	-	-	+	-	-	-	-	-	-	+	-	-	+	-	-	-	-	-	-	-	-
Lemna minor	-	-	+	+	-	-	-	-	-	+	+	-	-	+	-	-	-	-	-	-	+	-

Spirodela polyrhiza	-	-	+	-	-	-	-	-	-	+	-	-	-	-	-	-	-	-	-	-	-	-
Salvinia natans	-	-	-	+	-	-	-	-	-	-	-	-	-	+	-	-	-	-	-	-	-	-
Utricularia vulgaris	-	-	-	-	-	-	-	-	-	+	-	-	-	-	-	-	-	-	-	-	-	-
Hydrocharis morsus-ranae	+	-	+	-	-	-	-	-	+	-	-	-	-	-	-	-	-	-	-	-	+	-
Eupatorium cannabinum	-	+	-	-	+	-	+	-	+	1	1	2	-	+	-	-	+	+	-	-	-	
Bidens tripartitus	-	+	-	-	-	+	+	-	+	-	+	+	-	-	+	-	+	-	+	-	+	-
Urtica dioica	+	-	-	+	-	-	+	-	1	1	+	1	+	-	-	+	+	-	-	-	-	-
Pulicaria dysenterica	-	-	-	-	-	+	-	-	+	-	+	-	-	-	-	-	-	+	-	-	-	-
Polygonum hydropiper	-	+	-	-	+	-	+	-	+	+	-	-	-	+	-	-	-	-	-	-	-	+
Alopecurus ventricosus	-	-	-	-	-	-	-	-	-	-	-	-	+	-	+	-	+	-	-	-	-	-

Associations : 1 = *Scirpo – Phragmitetum* W. Koch 1926; 2 = subass. *salicetosum cinereae* Ştefan et al. 1995; 3 = *Thelypteridi – Phragmitetum* Kuiper 1958; 4 = *Typhetum angustifoliae* (All.1922)Pign. 1943; 5 = *Glycerietum maximae* Hueck 1931: 6 = *Phalaridetum arundinaceae* (W. Koch 1926) Libbert 1931; 7 = *Oenantho – Rorippetum* Lohmeyer 1950

References

Coldea, G., 1997. *Les associations végétales de Roumanie, tom 1, Les associations herbacées naturelles*, Presses Universitaires de Cluj.

Hanganu J., Dubyna D., Zhmud E., Grigoraş I., Menke U., Drost H., Ştefan N., Sârbu I., 2002. *Vegetation of the Biosphere Reserve Danube Delta – with Transboundary Vegetation Map on a 1:150.000 scale*. Danube Delta National Institute, Romania; M. G. Kholodny – Institute of Botany & Danube Delta Biosphre Reserve, Ukraine and RIZA, The Netherlands. RIZA rapport 2002.049, Lelystad.

Sanda, V., Popescu, A., 1973. Cercetări privind flora şi vegetaţia din Delta Dunării, *St. şi Cerc. de Biol., Seria Bot.*, **25** (5): 339-424.

Ştefan, N., Sârbu, I., Chifu, T. & Hanganu, J., 1995. Contribuţii la fitocenologia stufărişurilor din Delta Dunării, *Analele St. ale Inst. Delta Dunării*, **4** (1):179-199.

NEW OBSERVATIONS ON THE BIOLOGICAL FEATURES OF IZVORU MUNTELUI – BICAZ RESERVOIR

CARMEN AONCIOAIE[1], MIHAI-GEORGEL ERHAN[1] and MANUELA MIRON[2]
[1] "Alexandru Ioan Cuza" University of Iasi, Romania, Science Department: "Petre Jitariu" Biological Research Station, Potoci, Romania; e-mail: carmensilva2003@yahoo.com, ermikero@yahoo.com;
[2] "Alexandru Ioan Cuza" University of Iasi: "Petre Jitariu" Biological Research Station, Potoci, Romania, e-mail: mironmanuela@yahoo.com

Abstract. The paper presents the latest observations (August – December 2011) on the trophic state, phytoplankton and planktonic crustaceans composition of Izvoru Muntelui – Bicaz Reservoir in parallel with the data from a previous study (August – December 2009). While the 2009 studies presented the reservoir as oligo-mesotrophic with an yearly average of 2.6 µg.L^{-1} of chlorophyll-*a*, and the August/December average of 3.83 µg.L^{-1} of chlorophyll-*a,* the 2011 observations present a very different image, one of a meso-eutrophic reservoir with chlorophyll-*a* concentrations of 6.5 to 20.26 µg.L^{-1} (average of 12 µg.L^{-1}). Regarding the phytoplankton, in 2011 the effectives are higher than in 2009 as reflected in the trophy level. An interesting phenomenon is represented by the increase in Cyanophyta species and especially in effectives, due to an explosion of the autotrophic picoplankton colonies. The dominant diatom still remains *Cyclotella distinguenda* var. *unipunctata* and Bacillariophyta the main algal class of the reservoir. The epiphytic genus *Salpingoeca* (Choanoflagellata) is presented for the first time in the reservoir.Concerning the planktonic crustacean populations the total average densities are higher in 2009 than in 2011. Our analysis shows that the strong development of cyanophytes may be the reason of their decrease. The dominant species is represented by *Eudiaptomus gracilis*. Its average density in 2009 was of 971.1 ind. m^{-3} while in 2011 was of 899.3 ind.m^{-3}. The evolution of *E. gracilis* seem to be in accordance with the recorded trophic state, as scientific literature specify that it is a species that inhabits mostly a lower trophy level water (oligo- and mesotrophic). *Alona rectangula* and *A. gutatta* were identified only in the 2011 samples. *Acanthocyclops robustus* and *Simocephalus vetulus* are presented for the first time in the reservoir and *Leydigia leydigi* was again identified after many years.

Key words: Phytoplankton, autotrophic picoplankton, chlorophyll-*a*, biomass, planktonic crustacean populations, reservoir

Rezumat. Noi observaţii privind componentele biologice ale lacului de acumulare Izvoru Muntelui – Bicaz. Lucrarea prezintă cele mai noi observaţii (August – Decembrie 2011) asupra stării trofice, a compoziţiei fitoplanctonului şi populaţiilor de crustacee planctonice din lacul de acumulare Izvoru Muntelui – Bicaz, în paralel cu datele obţinute pe parcursul unui studiu anterior (August – Decembrie 2009). În timp ce studiile din 2009 prezentau lacul de acumulare ca fiind oligo-mezotrof cu o medie anuală a clorofilei-*a* de 2,6 µg.L^{-1} şi o medie August/Decembrie de 3,83 µg.L^{-1}, observaţiile din 2011 prezintă o situaţie foarte diferită, cea a unui lac mezo-eutrof cu o serie de concentraţii de clorofilă-*a* între 6,5 şi 20,26 µg.L^{-1} (concentraţie medie: 12 µg.L^{-1}). În ceea ce priveşte fitoplanctonul, efectivele identificate în 2011 sunt mai mari decât în 2009, ceea ce se reflectă în starea trofică. Un fenomen interesant este reprezentat de creşterea în numărul de specii dar mai ales creşterea numerică a algelor din clasa Cyanophyta datorită unei explozii a coloniilor de picoplankton autotrof. Diatomeea dominantă rămâne încă *Cyclotella distinguenda* var. *unipunctata* iar Bacillariophyta, clasa de alge predominantă în acest lac de acumulare. Genul epifitic *Salpingoeca* (Choanoflagellata) este identificat pentru prima dată în lac. În ceea ce priveşte populaţiile de crustacee planctonice, densitatea totală medie este mai mare în 2009 decât în 2011. Analizele noastre arată că această descreştere pare a se datora dezvoltării puternice a cianofitelor. Specia dominantă este reprezentată în ambele perioade analizate de către *Eudiaptomus gracilis*. Densitatea sa medie a fost în 2009 de 971,1 ind. m^{-3} şi în 2011 de 899,3 ind.m^{-3}. Evoluţia lui *E. gracilis* pare a fi în concordanţă cu starea trofică din perioadele de timp luate în studiu, deoarece literatura stiinţifică specifică faptul că această specie se dezvoltă mai ales în ape cu troficitate redusă (oligo- şi mezotrofe). Speciile *Alona rectangula* şi *A. gutatta* au fost identificate doar în probele colectate în 2011, nu şi în 2009. *Acanthocyclops robustus* şi *Simocephalus vetulus* sunt prezentate pentru prima dată în lac iar *Leydigia leydigi* este regăsită după mulţi ani.

Cuvinte cheie: Fitoplancton, picoplancton autotrof, clorofila-*a*, biomasă, populații de crustacee planctonice, lac de acumulare

Introduction

The objective of the present study is to point out the current situation of the biological features of Izvoru Muntelui – Bicaz Reservoir in a five months period (August – December 2011) in parallel with the previous data recorded in 2009. The study regards the change in trophy level and the modifications that occurred in the composition of phytoplankton and planktonic crustacean populations. The study also follows the physico-chemical characteristics of the water column.

Material and Methods

Study area. Izvoru Muntelui – Bicaz Reservoir, located in Eastern Romania on Bistrița River (Oriental Carpathians) had as building purpose the necessity of storing a large quantity of water as supply for the hydro-electrical power plant build at Stejaru (Neamț County). The geographical coordinates are the following: 47º05'39" N and 25º57'18" E in the upper region (at maximum length and full volume) and 46º 56'18" N and 26º06'10" E at the dam (Google Earth). The maximum depth, reached at the dam, varies between 75 and 90 m and, at maximum volume, the reservoir contains 1.23 x 10^9 m^{-3} of water (Miron *et al.*, 1983). The reservoir is thermally stratified and dimictic (Miron *et al.*, 1983). The trophic state of the reservoir varied during the years from oligotrophic to eutrophic.

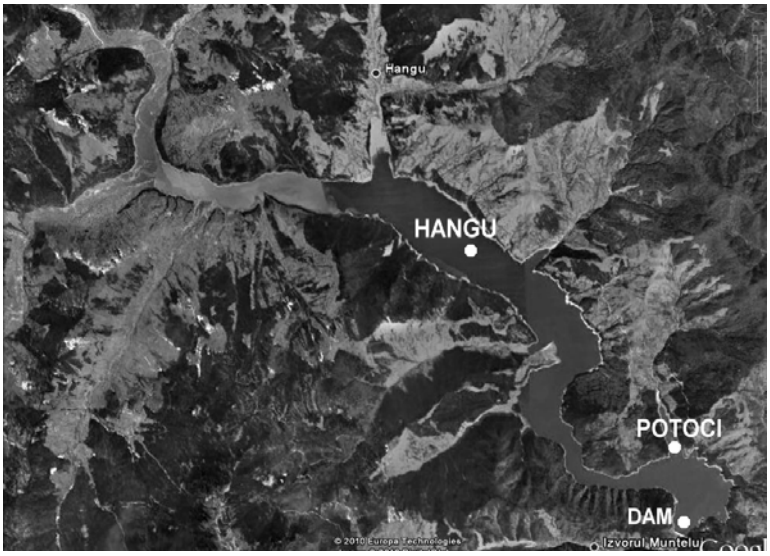


Figure 1. Izvoru Muntelui – Bicaz Reservoir: location of the three sampling points. Source: Google Earth (14.09.2010).

Methods.

Sampling took place once a month from August to December 2009/2011 in three sampling points located in key regions of the reservoir (a gulf area – Potoci Gulf; the dam area and the middle of the reservoir – Hangu). Sampling consisted in collecting water samples, measuring the Secchi depth and recording the physico-chemical data. The physical and chemical analyses were made with the help of the Multiparameter Sonde Type 6600 V2-2 and the recorded parameters were depth, temperature, dissolved oxygen (D.O.), pH, O.R.P. and conductivity. The photic zone depth was calculated by multiplying the Secchi disk depth with the 2.5 factor (Surugiu, 2007).

For the phytoplankton analysis, the samples were collected with a Wildco Van – Dorn horizontal water sampler (volume: 2.2 liters) from 3 depths (0; 5 and 10 m). 1 liter of water from each depth was stored in a clean plastic recipient and preserved at site with 5 ml of 37% formaldehyde and Lugol solution. In the laboratory, the samples were let to sediment for two weeks and the sediment was stored in special recipients. For diatoms, the samples were mineralized and mounted in a high refractive index medium (Karthick *et al.*, 2010). The quantitative analysis of the phytoplankton taxa was made using the Utermöhl method (Lars & Elbrächter in Karlson *et al.*, 2010, Chapter 2: 13 - 20).

Chlorophyll-*a* concentration ($\mu g.L^{-1}$) was determined by spectrometric method (standardized method SR ISO 10260 Romanian Standard) and the trophic classes assessed after Ryding & Rast (1994). The phytoplankton biomass was calculated using chlorophyll-*a* concentration multiplied with the factor 67 (Surugiu, 2007).

Planktonic crustaceans' samples were collected using a Birge closing net (mesh size 80 μm) for the quantitative analysis and a simple planktonic net with the mesh size of 100 μm for qualitative analysis. Samples were preserved at site with 96% ethanol. Laboratory stage was represented by the concentration of samples, identification of planktonic crustacean species and counting of the individuals using specific procedures involving dissection, binocular and microscope analysis.

Results and discussions

The present study presents the differences regarding the biological features of Izvoru Muntelui – Bicaz Reservoir that occurred since 2009 in parallel with the data obtained during a study of five month in 2011 (August – December). The reservoir was analyzed from the angle of physico-chemical data, chlorophyll-*a*, phytoplankton biomass, qualitative and quantitative phytoplankton and zooplankton analysis.

Physico-chemical analysis

The analysis of the physical and chemical characteristics of the reservoir was made along the water column from the surface to 40 m, covering the depths where the biological samples were collected. The transparency (Secchi disk depth) varied in 2009 from 1.75 to 4.33 m (3.12 m average) and in 2011 from 2.9 to 5.23 m (4.21 m average). The photic zone thus was deeper in 2011 (10.5 m average; min. 7.25 m; max. 13.1 m) than in 2009 (7.8 m average; min. 4.37 m; max. 10.83 m). In all analyzed months the transparency was higher in 2011. Similar values of the transparency occurred only in November and December 2009/2011.

The graphics bellow (Fig. 2 and 3) present the water characteristics in parallel for each month, side by side 2009 and 2011, for a clearer view of the resemblances or differences. Regarding the temperatures (fig. 2A), the differences are not high; a note may be made on the fact that in August and September 2011 was warmer than in 2009 by at least 1ºC, while in the rest of the months it was colder in 2011 than in 2009 but with small differences (October and November: 1.2 / 0.6ºC; December: 1.39ºC).

At the surface the dissolved oxygen concentration ($mg.L^{-1}$) inversely correlates with the temperature, as in August and September 2011 was smaller than in 2009 while for the rest of the months was higher. An oxygen decrease at certain depths was previously noticed in 2009, phenomenon that maintained in 2011 with the specification that in September 2011 the concentration strongly fluctuated first decreasing in the 5-10 m interval than again after 15 m. This phenomenon, as seen in figure 2B, occurred during the warmer months 2009/2011. In October 2009/2011 the decrease begins around 25 m depth and under 30/35 m for November and December 2009/2011. Except November and December 2009/2011, the oxygen concentrations begin to increase again around 35-40 m depth.

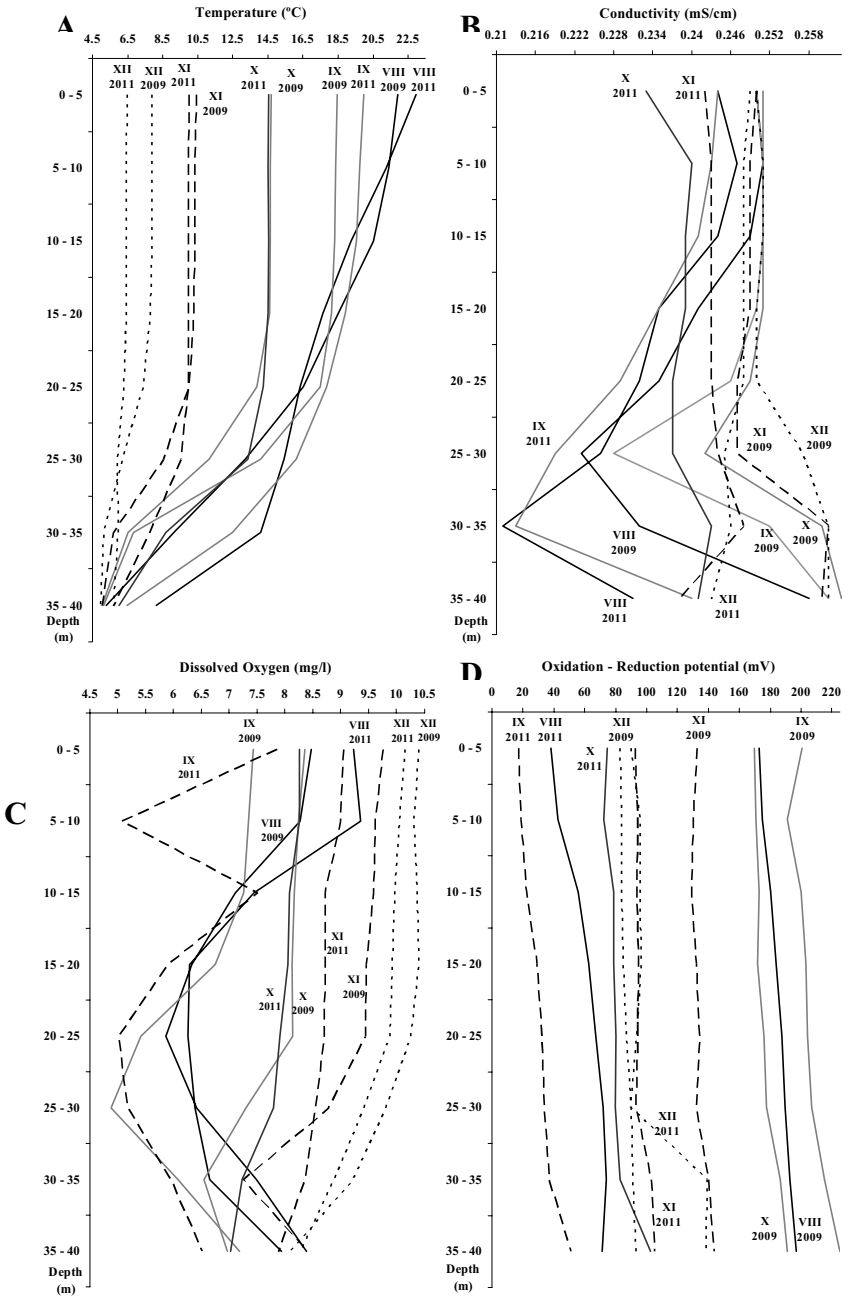


Figure 2. Physico-chemical properties (**A**: Temperature, **B**: Dissolved Oxygen, **C**: Conductivity, **D**: Oxidation – Reduction Potential).

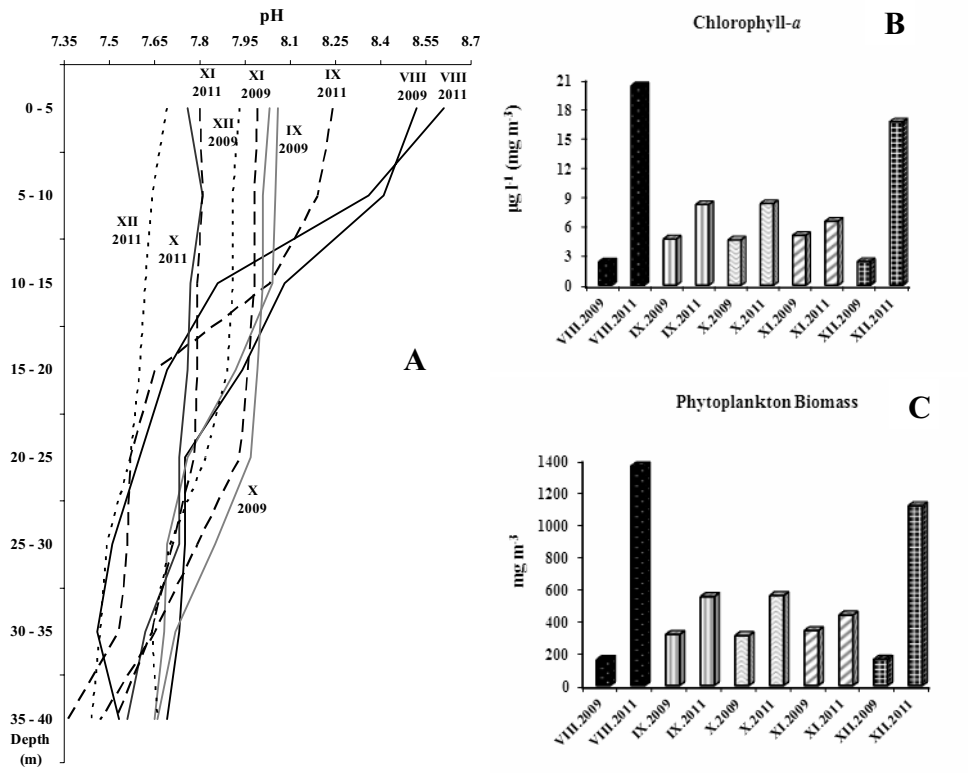


Figure 3. Physico-chemical properties (**A**: pH);
Biological characteristics (**B**: Chlorophyll-*a*, **C**: Phytoplankton biomass).

Conductivity (mS/cm) has almost the same values in 2009 and 2011. As seen in figure 2C each month follows a similar pattern no matter the year, even if the values may decrease/increase more or less.

The O.R.P. (Oxidation – Reduction potential, mV) shows a definite difference between 2009 and 2011. The year 2009 in characterized by high values in the 83-225 mV interval (mostly above 130 mV), while 2011 around 17-138.8 mV but mostly under 100 mV (Fig. 2D).

The pH varies from 7.47 to 8.52 in 2009 and from 7.36 to 8.61 in 2011 (Fig. 3A). An observation is that pH is positively correlated to temperature as in August and September 2011 was higher than in 2009 and in the other three month was lower (Fig. 2A). In august 2011, at around 5-10 m it can be noticed the same decrease in pH as it is in temperature (Figs. 2A/3A).

In 2009, the ammonium (NH_4^+) analysis showed an average of 0.26 $mg.L^{-1}$, for nitrate (NO_3^-) of 1.03 $mg.L^{-1}$ and for phosphorous of 0.042 $mg.L^{-1}$ (Miron *et al.*, 2010). In 2011 the ammonium, nitrate and nitrite had barely identifiable traces while phosphorous was higher ≈ 0.15 $mg.L^{-1}$. In 2011 water hardness was also assessed. Carbonate hardness had an average of 6.9ºd and the ABC (acid binding capacity) of 2.6 $mmol.L^{-1}$; total hardness had an average of 8.2ºd and the calcium carbonate

($CaCO_3$) average concentration was 151.9 $mg.L^{-1}$; the residual hardness was constant around 0.5ºd/ 0.6ºe.

Biological features analysis

The trophicity of the reservoir was assessed since the begging of the reservoir, and during the years was noticed an increase from mesotrophy to eutrophy (1969–1983).

In 1963, after its formation, the reservoir was presented as oligotrophic (Popescu & Oltean, 1963). First tendency to mesotrophy was noticed in 1964 when *Rhizosolenia longiseta* presented a strong bloom, phenomenon that appeared again in 1965 (1600 – 40000 x 10^3 ex. L^{-1}) according to Cărăuş (1969). The species lasted two more years in the reservoir and disappeared until identified by Darabă (2008a). In 1969, the increase in trophy level was shown by blooms of *Oscillatoria rubescens* (3280 x 10^3 ex. L^{-1}), blooms that occurred in the following years along with blooms of *Scenedesmus* and *Dinobryon sociale* (Cărăuş, 1970; 1979; Miron *et al.*, 1983). Also, the appearance and increase in species characteristic to mesotrophic and eutrophic waters, and ponds (*Micractinium bornhemiense*, *Golenkinia radiata* and others) showed the increase in trophy level (Miron *et al.*, 1983). This increase is clearly specified in Cărăuş (1979) when, after 13 years of continuous studies, the jump from 3 $g.m^{-3}$ to 10 $g.m^{-3}$ of phytoplankton total biomass, shown first a transition from mesotrophy to eutrophy and finally the installation of eutrophy. From 2007 to 2010 the lake was presented as oligo-mesotrophic (Daraba, 2008b; Aoncioaie & Erhan, 2010; Miron *et al.*, 2010) with strong variations from ultraoligotrophy to mesotrophy but not above. The latest data (present study) present the reservoir as meso-eutrophic. The differences among the chlorophyll-*a* concentrations from 2009 and 2011 can be seen in figure 3B. The average chlorophyll-*a* concentration in 2009 (August - December) was 3.83 $\mu g.L^{-1}$ with values from as low as 2.4 $\mu g.L^{-1}$ to the highest of 5 $\mu g.L^{-1}$ while in 2011, the average concentration grew to 12 $\mu g.L^{-1}$ due to values from 6.5 $\mu g.L^{-1}$ to 20.3 $\mu g.L^{-1}$. In correlation with chlorophyll-*a* concentration, the phytoplankton biomass is also higher in 2011 in comparison to 2009 (Fig. 3C). The average biomass in 2009 (August – December) was 256.6 $mg.m^{-3}$ and in 2011 (same period) was 804 $mg.m^{-3}$. The highest biomass identified was of 1357.4 $mg.m^{-3}$ in August 2011, followed by December 2011 with a biomass of 1,112.2 $mg.m^{-3}$ while in 2009 the highest biomass was only of 339 $mg.m^{-3}$ in November.

Phytoplankton analysis

The phytoplankton qualitative analysis shows a number of 7 classes of which Choanoflagellata (represented by *Salpingoeca* spp.) was not identified in 2009. Quantitatively speaking, the year 2011 is 7 times richer in ex. L^{-1} then 2009 (phytoplankton average in the study period: 14,542.63 x 10^3 ex. L^{-1} in 2011 and 1,976. 9 x 10^3 ex. L^{-1} in 2009).

Bacillariophyta remains the dominant class in Izvoru Muntelui – Bicaz Reservoir and the diatom effectives are higher in 2011 than in 2009 for the studied months (Fig. 4A). The average diatoms density was 1,931 x 10^3 ex. L^{-1} in 2009 and 12,344.84 x 10^3 ex. L^{-1} in 2011 for August to December. Even though, the diatoms species list is smaller in 2011 than in 2009. Species like *Cymatopleura elliptica*, *C. solea*, *Diatoma mesodon*, *Didymosphaenia geminata*, *Gyrosigma attenuatum*,

Neidium dubium, *Pinnularia microstauron*, *Stauroneis anceps*, *S. phoenicenteron*, *Stephanodiscus astraea*, *Synedra capitata* etc. that occured frequently in 2009 were not yet identified. *Cyclotella distinguenda* var. *unipunctata* remains the dominant diatom.

Regarding all other 6 algal classes the situation also changes radically from 2009 to 2011 and next to the increase/decrease fluctuation in species number and in their densities, a great amount of autotrophic picoplankton from Cyanophyta (unidentified before) bloomed in October and December 2011 (Fig. 4B). The average density of cyanophytes was 1.25 x 10^3 ex. L^{-1} in 2009 and 1,746.36 x 10^3 ex. L^{-1} in 2011. The species number is higher in 2011 to the previous two identified in 2009 (*Merismopedia elegans* and *Oscillatoria rubescens*) adding the colonies of autotrophic picoplankton, *Aphanothece, Rhabdoderma, Synecoccocus* and species like *Chroococcus turgidus*, *Microcystis aeruginosa*, *Oscillatoria limnetica*, *O. limosa*.

In correlation with the chlorophyll-*a* concentration it can be seen that in August and December 2011 there are the highest trophy levels due to the high density of Bacillariophyta in August 2011 and of Cyanophyta plus Dinophyta and Euglenophyta in December 2011. The concentration of chlorophyll-*a* in October 2011 is not high, despite of the highest density of autotrophic picoplankton, due to a very small development of the rest of the phytoplanktonic classes. The importance of the autotrophic picoplankton is underlined by Malinsky-Rushansky et al., 1997 that specifies how in certain conditions of light, depth and abundance it could be responsible for most of the photosynthetic activity.

Chlorophyta class had a higher density in August 2009 than in August 2011, and almost disappeared in September, October and December as it occurred in extremely small effectives. In 2011 occurred constantly each month with a peak in September. The average density was 40.8 x 10^3 ex. L^{-1} in 2009 and 288.51 x 10^3 ex. L^{-1} in 2011 for the five studied month (Fig. 4C). A higher number of species were identified in 2011 among which *Ankyra judayi*, *Ankistrodesmus gracilis*, *Dactylococcopsis acicularis*, *Elakatothrix spirochroma*, *Gonatozygon brebissonii*, *Monoraphidium contortum*, *M. griffithii*, *Siderocystopsis fusca*, *Scenedesmus ecornis*, *S. linearis*, and *Treubaria triappendiculata* not identified in 2009.

Dinophyta (Fig. 4D) also had smaller average densities in 2009 (3.62 x 10^3 ex. L^{-1}) than in 2011 (21.81 x 10^3 ex. L^{-1}). The species were the same in both studied periods. Euglenophyta class (Fig. 4E) was much more numerous in 2011 (average in 2009 of 0.12 x 10^3 ex. L^{-1} and 9.1 x 10^3 ex. L^{-1} in 2011). In 2011 the identified species occurred in a more uniform pattern than in 2009 during the studied months. Chrysophyta class (Fig. 4F) was much more numerous in 2011 (average in 2009 of 0.087 x 10^3 ex. L^{-1} and 22.71 x 10^3 ex. L^{-1} in 2011). Choanoflagellata (Fig. 4G) was newly identified in Izvoru Muntelui – Bicaz Reservoir in the form of the epiphytic genus *Salpingoeca* that coexists with and depends on *Asterionella formosa* (2009: 0 ex. L^{-1}; 2011: 109.3 x 10^3 ex. L^{-1}).

Planktonic crustacean populations analysis

The taxonomical list presents, next to the common ones, five species that were not identified in 2009 (*Alona rectangula*, *A. costata*, *Acanthocyclops robustus, Leydigia leydigi, Simocephalus vetulus*). *Alona guttata* was, next to the previous

ones, not identified in 2010 samples. *A. robustus* and *S. vetulus* are presented for the first time in the reservoir and *Leydigia leydigi* is reconfirmed after many years (Miron *et al.*, 1983).

Regarding the total average densities, the general image that can be seen in Fig. 5A is that in 2009 the number of planktonic crustaceans was definitely higher than in 2011. The average density was 1,903.6 ind. m^{-3} in 2009 and 287.3 ind. m^{-3} in 2011. This is mainly due to high densities identified in August 2009 in all sampling stations (3,920.8 ind. m^{-3}) and in Hangu sampling station also in September (4,228.9 ind. m^{-3}) and December (3,054.8 ind. m^{-3}) 2009. In the other two sampling stations (Dam area and Potoci), from September to December 2009 the values were lower but still higher than the ones identified in 2011 – same period.

The dominant planktonic crustaceans are represented in 2009 and 2011 by *Eudiaptomus gracilis*, *Diaphanosoma orghidani*, *Daphnia cucullata* and *D. galeata*. Samples collected in August 2010 show the presence of the same species as dominant in the spectrum but in lower densities (Fig. 5B). *E. gracilis* and *D. orghidani* had the maximum densities in 2009 and the minimum in 2010, in 2011 the densities being around and above half of the 2009 average densities. In the case of both *Daphnia* species, the minimum values were counted in 2011 while the maximum for *D. galeata* was identified in 2009 (116 ind. m^{-3}) and for *D. cucullata* in 2010 – 276.9 ind. m^{-3} (Fig. 5B).

A much more detailed analysis for *E. gracilis* (Fig. 5C), which is the dominant crustacean in Izvoru Muntelui – Bicaz Reservoir, shows that its densities fluctuate between 2009 and 2011 and that the difference is made by August and December 2009 when its densities were higher than 2011. In September, October and November 2011 the average densities were higher than in 2009, same months. The average density was 971.1 ind. m^{-3} in 2009 and of 899.3 ind.m^{-3} in 2011. The evolution of the *E. gracilis* densities seems to be in accordance with the recorded trophic state (2009 oligotrophy, 2011 mesotrophy), as scientific literature specify that it is a species that mostly inhabits a lower trophy level water. Still, the newly identified trophic state is not reflected in the development of species like *Chydorus sphaericus* and *Bosmina longirostris* which, according to the literature, are characteristic to high trophic level waters.

D. orghidani is also one of the dominant species in Izvoru Muntelui – Bicaz Reservoir although not reaching such densities as *E. gracilis*. In 2009, the general average was of 453.6 ind. m^{-3} and in 2011 of 213.7 ind. m^{-3} and. In all analyzed months the densities were higher in 2009.

If correlating the planktonic crustaceans densities in 2011 with the densities recorded for the phytoplankton it can be noticed that is negatively correlated. The newcomers in the phytoplankton community are mainly the autotrophic picoplankton colonies that belong to Cyanophyta and the scientific literature specifies that even if cyanophytes are ingurgitated by zooplankton, most of the times they pass through their gut undigested and have low-nutrition levels (Quiblier, 1994; Haney, 1987). Even if consumed, the dimensions and shape oh the cyanophytes is important, as smaller ones are preferred instead of colonial and filamentous (Thys, 2003). At the same time, even if consumed, cyanophytes are avoided while blooming (Thys, 2003) due to the secretion of allelopathic substances that are meant

to inhibit the development of the rest of phytoplankton and also due to the secretion of toxins (Haney, 1987). Another reason in planktonic crustaceans decrease may be as an indirect result created by the direct competition between cyanophytes and the rest of the phytoplankton, whose growth is inhibited by allelopathic substances (Haney, 1987). Literature specifies that when *Oscillatoria* and other cyanophytes are dominant, extremely low densities of cladocerans were observed and usually, the rotifers are positively correlated with the development of cyanophytes as the small-bodied rotifers may be less susceptible to cyanophyte toxin (Beaver, 1999).

The effect of phytoplankton on zooplankton is visible in October 2011 when the Cyanophyta class had a strong development that started in September, and even inhibited the other algal classes (Fig. 4 A – G) not only the planktonic crustaceans' populations. In figure 5A can be seen that in October 2011 the planktonic crustaceans had the lowest densities recorded in the study period from 2011, followed by December 2011 when the cyanophytes grew again in density but not as much as to strongly inhibit the other algal classes, enough though to inhibit again the development of planktonic crustaceans.

Conclusions

After a close analysis we reached the conclusion that the extreme increase in trophic state of Izvoru Muntelui – Bicaz Reservoir in 2011 represents a consequence of a powerful development of the algal classes mainly to the cyanophytic autotrophic picoplankton, related also to their more uniform and constant development month after month. Except Bacillariophyta class, which had less species than previously identified, more species were encountered for Cyanophyta, Chlorophyta and Choanoflagellata. The total average density of planktonic crustaceans was higher in 2009 than in 2011. The reason that the planktonic crustaceans are less in numbers in 2011 while the phytoplankton has greater effectives may be due to the cyanophytes development that have a direct effect through the low nutrition level and the inhibiting effect of the toxins comprised by the members of Cyanophyta class, and an indirect effect due to the inhibition of the phytoplankton development due to the secretion of allelopathic substances. *Eudiaptomus gracilis* development is in accordance with the recorded trophic state but the newly identified higher trophic state is not reflected in the development of species like *Chydorus sphaericus* and *Bosmina longirostris* which, according to the literature, are characteristic to high trophic level waters.

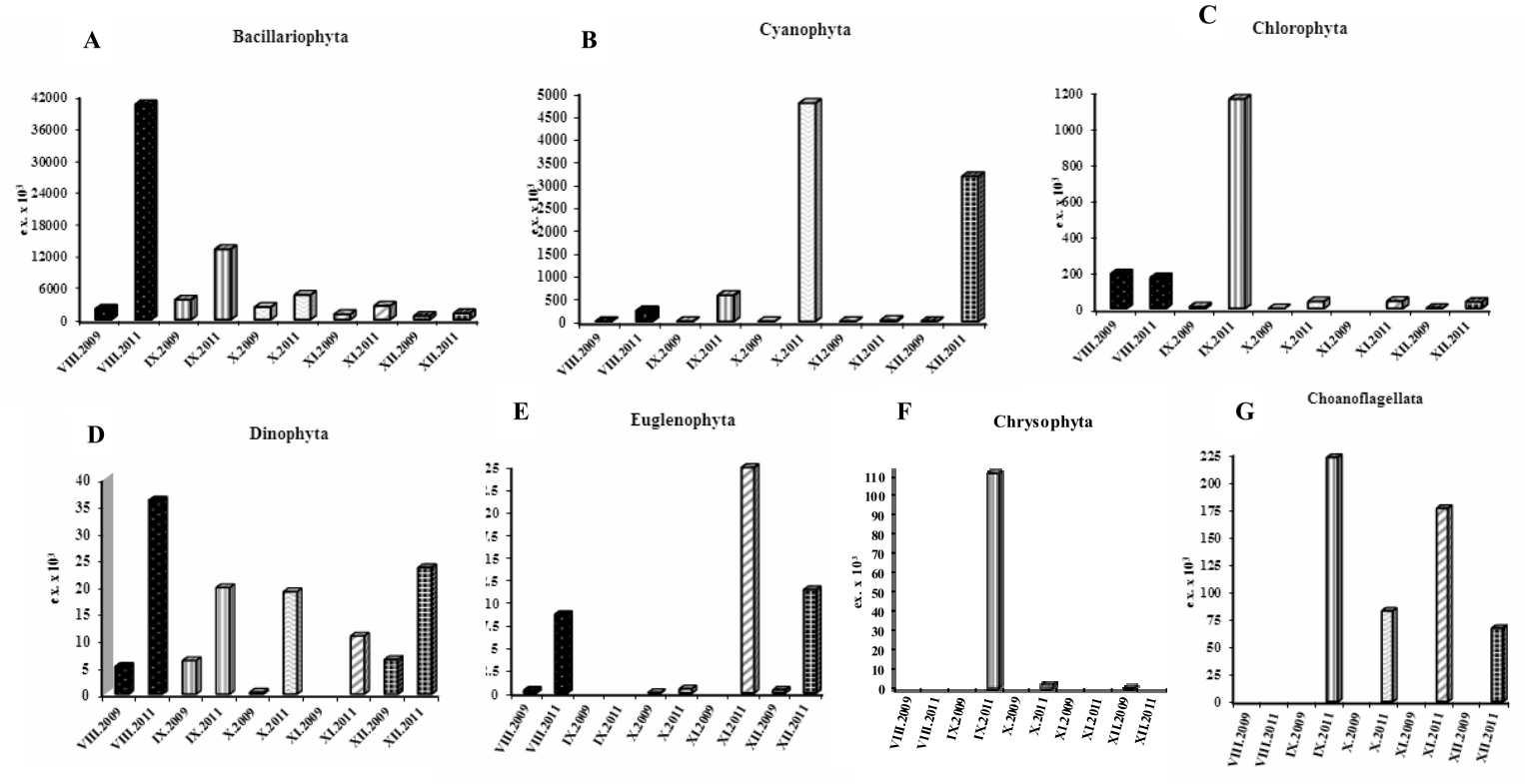


Figure 4. Phytoplanktonic classes and the algal densities in August – December 2009/2011.

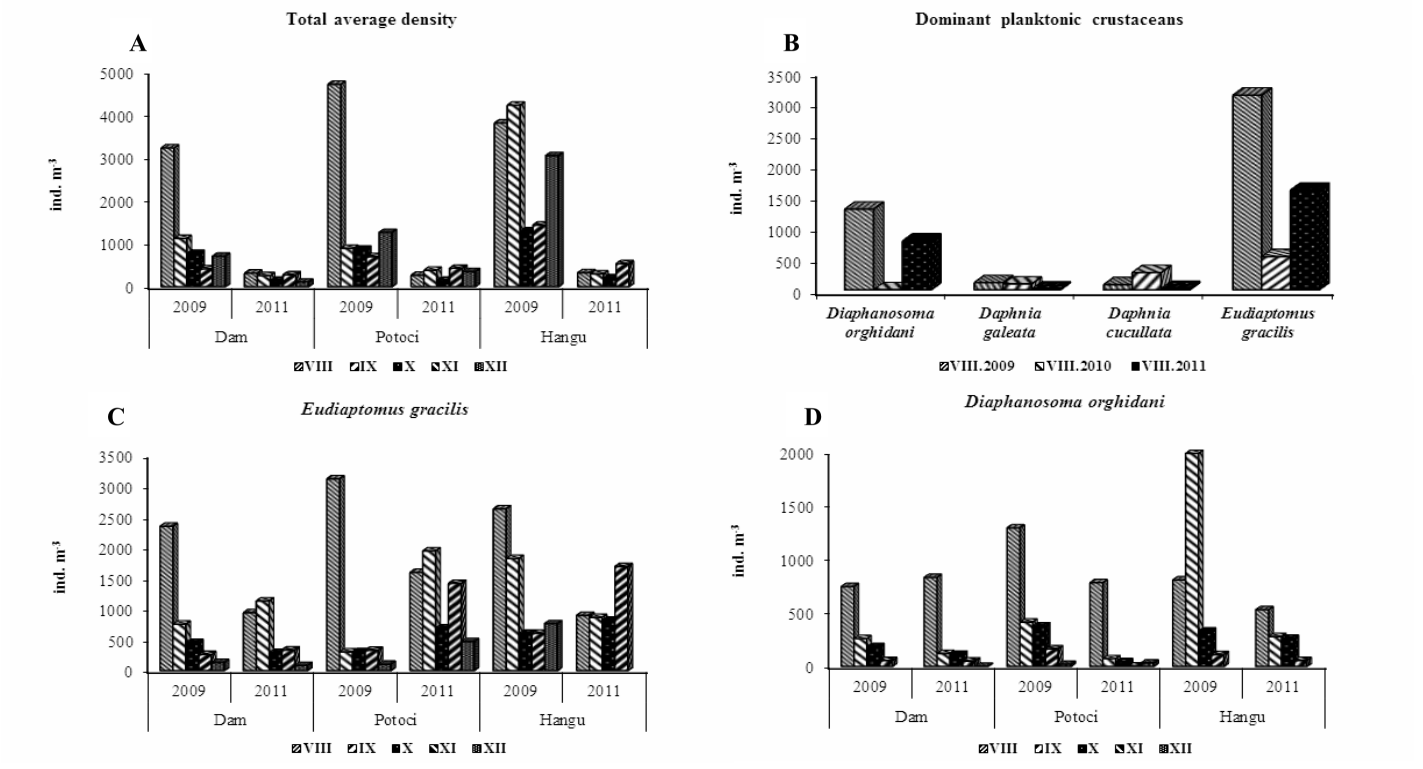


Figure 5. A: Planktonic crustacean populations total average densities in August – December 2009/2011; **B**: Density comparison among the dominant species in 2009, 2010 and 2011; **C**: *Eudiaptomus gracilis* densities in 2009/2011; **D**: *Diaphanosoma orghidani* densities in 2009/2011.

References

Aoncioaie, C., Erhan, M. G., 2010. Evaluation of the Bicaz Reservoir trophic state using *chlorophylla* as indicator. *Analele Ştiinţifice ale Universităţii "Al. I. Cuza" Iaşi, s. Biologie Animală*, **LVI**: 237–247.

Beaver, J. R., Miller-Lemke, A. M., Acton, J. K., 1999. Midsummer zooplankton assemblages in four types of wetlands in the Upper Midwest, USA. *Hydrobiologia*, 380: 209–220.

Callieri C., Stockner J. G., 2002. Freshwater autotrophic picoplankton: a review. *Journal of Limnology*, **61** (1): 1–14.

Cărăuş, I., 1969. The phytoplankton of Izvoru Muntelui – Bicaz water reservoir (II). *Lucrările Staţiunii de Cercetări Stejarul*, **2**: 152–162.

Cărăuş, I., 1970. Some data on the horizontal microdistribution of the phytoplankton in Izvoru Muntelui – Bicaz water reservoir: a methodological interpretation (I). *Lucrările Staţiunii de Cercetări Stejarul*, **3**: 251–257.

Cărăuş, I., 1979. The quantitative dynamics of the phytoplankton of Izvoru Muntelui – Bicaz water power reservoir (Romania): 1966 – 1978. *Travaux de Station Stejarul, Limnologie*, **7**: 97–110.

Darabă, O. M., 2008a. Distribuţia spaţio – temporală a diatomeelor (Bacillariophyceae) din planctonul lacului Izvoru Muntelui – *Izvoru Muntelui – Bicaz*; Ed. PIM Iaşi.

Darabă, O. M., 2008b. Trophic level of the Izvoru Muntelui – Bicaz Reservoir usng the chlorophyll a concentration as indicator, according to the Romanian Standard S.R. ISO 10260/1997. *Analele Ştiinţifice ale Universităţii "Al. I. Cuza" Iaşi, s. Biologie Animală*, **LIV**: 135–143.

Haney, J. F., 1987. Field studies on zooplankton – cyanobacteria interactions. *New Zealand Journal of Marine and Freshwater Research*, **21**: 467–475.

Hillbrand, H., Dürselen, C. D., Kirschtel, D., Pollingher, U., Zohary, T., 1999. Biovolume calculation for pelagic and benthic microalgae. *Journal of Phycology*, **35**: 403–424.

Hutorowicz, A., 2005. Opracowanie standardowych objętości komórek do szacowania biomasy wybranych taksonów glonów planktonowych wraz z określeniem sposobu pomiarów i szacowania. Instytutu Rybactwa Śródlądowego.

Karlson, B., Cusak, C., Bresnan, E., 2010. *Microscopic and molecular methods for quantitative phytoplankton analysis. Intergovernmental Oceanographic Commission. Manuals and Guides. UNESCO*.
Available from http://www.jodc.go.jp/info/ioc_doc/Manual/187824e.pdf (access date: 2.IX.2010)

Karthick, B., Taylor, J. C., Mahesh, M. K., Ramachandra, I. V., 2010. Protocols for collection, preservation and enumeration of Diatoms from aquatic habitats for water quality monitoring in India. *The Icfai. University Journal of Soil and Water Sciences*, **III** (1): 25–60.

Malinsky-Rushansky, N. Z., Berman, T., Dubinsky, Z., 1997. Seasonal photosynthetic activity of autotrophic phytoplankton in Lake Kinneret, Israel. *Journal of Plankton Research*, **19** (8): 979–993.

Miron, I., Cărăuş, I., Măzăreanu, C., Apopei, V., Grasu, C., Ichim, I., Mihăilescu, F., Rujinschi, R. I., Rujinschi, C., Simalcsik, F., Misăilă, C., Ţăruş, T., Apetroaiei, N., 1983. *Lacul de acumulare Izvoru Muntelui – Bicaz. Monografie Limnologică.* Ed. Acad. R.P.R. Bucureşti.

Miron, I., Neagu, A. N., Erhan, M. G., Aoncioaie, C., Fusu, M. M., 2010. *Succesiunea ecologică. Râul Bistriţa – Lacul Bicaz. Monografie limnologică II*. Ed. Univ. „Al. I. Cuza" Iaşi.

Popescu, E., Oltean, M., 1963. Dinamica rezervelor trofice planctonice din lacul de acumulare Bicaz, în primii doi ani de la inundare. Buletinul Institutului de Cercetări şi Proiectări Piscicole, **XXII** (2): 32–50

Quiblier, C., Bourdier, G., Amblard, C., Pepin, D., 1994. Separation of phytoplanktonic pigments by HPLC for the study of phyto-zooplankton trophic relationships. *Aquatic Sciences*, **56**/1: 29–39

Romanian Standard S.R. ISO 10260, 1997. Water Quality. Measurement of biochemichal parameters. Spectrometric determination of chlorophyll-a concentration.

Ryding, S. O., Rast, W., 1994. *Le contrôle de l'eutrophisation des lacs et des réservoirs*. Ed. Masson Paris.

Surugiu, V., 2007. *Ecologie Marină. Îndrumar pentru lucrări practice*. Ed. Univ. "Al. I. Cuza" Iaşi.

Thys, I., Leporq, B., Descy, J. P., 2003. Seasonal shifts in phytoplankton ingestion by *Daphnia galeata*, assessed by analysis of marker pigments. *Journal of Plankton Research*, **25**: 1471–1484

Wetzel, R. G., 2001. *Limnology: Lake and river ecosystems*. Academic Press, San Diego.

FERTILITY EFFECTS ON *ULVA* THALLI MASS DEVELOPMENT IN INLAND WATERS OF POLAND

BEATA MESSYASZ, ANDRZEJ RYBAK, MARTA PIKOSZ and LIDIA SZENDZINA

Department of Hydrobiology, Institute of Environmental Biology, Adam Mickiewicz University, Umultowska 89, 61-614, Poznań, Poland, e-mail: messyasz@amu.edu.pl

Abstract. The genus *Ulva* (Ulvophyceae, Chlorophyta) consists of some 140 species which are present mainly in marine waters. Nine species occur in the inland waters in Poland (central Europe), of which *U. flexuosa* subsp. *pilifera* (Kützing) Bliding is the most widespread in the Wielkopolska region. The occurrence of *Ulva* species in any particular locality seems to be governed mainly by the availability of nutrients (N, P) than by the high levels of anthropogenic salinity in freshwaters. Freshwater *Ulva* can grow in different types of ecosystems from reservoir (Malta) through ponds (Tulce) or rivers (Nielba) and ending on drainage ditches. The main question addressed by this study was does the quantity and size of *Ulva* thalli increase together with the amount of nutrients in water? In addition, the aim of the study was to determine whether the development and the persistence for the long time of the big and dense *Ulva* patches exert influence for the aqueous ecosystem. We used freshwater *Ulva* as a model study species.

Key words: macroalge, *Ulva* spp., freshwater ecosystems, thallus and cells size, morphology, Poland

Rezumat. Efectele fertilității asupra dezvoltării în masă a talului de *Ulva*, in apele interioare ale Poloniei. Genul *Ulva* (Ulvophyceae, Chlorophyta) este format din aproximativ 140 specii prezente în principal în apele marine. Nouă specii apar în apele interioare în Polonia (Europa Centrală), din care *U. flexuosa* subsp. *pilifera* (Kützing) este cea mai răspândită în regiunea Wielkopolska. Apariția speciilor de *Ulva*, pare a fi reglementată mai mult de disponibilitatea de elementelor nutritive (N, P), decât de un nivel ridicat al salinității din cauze antropice, în apele dulci. Speciile de *Ulva* de apă dulce pot să crească în diferite tipuri de ecosisteme, de la baraje (Malta), bălți (Tulce) sau râuri (Nielba) și până la șanțuri de drenaj. Scopul acestui studiu este de a vedea în ce măsură cresc cantitatea și dimensiunea talului la *Ulva* în funcție de aportul de nutrienți. În plus, scopul studiului a fost de a determina dacă dezvoltarea și persistența pentru mult timp a câmpurilor dense de *Ulva* exercită vreo influență pentru ecosistemele acvatice. Ca model de studiu am utilizat o specie de *Ulva* de apă dulce.

Cuvinte cheie: macroalge, *Ulva* sp., ecosisteme acvatice dulcicole, dimensiunea celulară și a talului, morfologie, Polonia

Introduction

The genus *Ulva* (Ulvophyceae, Chlorophyta) including species previously classified as the genus *Enteromorpha* (Hayden *et al.*, 2003) is frequent macroscopic green algae of the marine littoral zone and estuaries - widely distributed all over the world (Bäck *et al.*, 2000; Kirchhoff & Pflugmacher, 2002; Apeng *et al.*, 2008) and often shows a tendency to form blooms (Marěs *et al.*, 2011). Since the nineteenth century, occasionally, *Ulva* thalli been observed in the inland waters of Poland, mainly in northern and western parts of the country (Kozłowski, 1890; Sitkowska, 1999). However, in recent years their presence has significantly increased – particularly in Wielkopolska region (Messyasz & Rybak, 2009a). Currently recognize in Polish Baltic Sea thirteen species (Pliński & Jóźwiak, 2004) and nine species in inland waters (Rybak & Messyasz, 2009). The spread of marine *Ulva*

species into freshwater environment causes disturbances in structure and functioning of ecosystems.

In the Mondego estuary (west Portugal) growth rates of the opportunistic macroalgae – *Ulva intestinalis* L. (syn. *Enteromorpha intestinalis*) primarily depends on salinity (Martins *et al.*, 1999). However, the dominance of *Ulva* species in any particular locality in freshwaters seems to be mainly governed by the availability of nutrients, especially nitrogen and phosphorus than by the high levels of anthropogenic salinity (Messyasz & Rybak, 2010). Nutrient enrichment can cause change habitat of ecosystems and lead to excessive growth of primary producers (Zheng & Paul, 2012).

Ulva can colonize lakes as well as ponds or rivers. It also occasionally grows in very slowly moving shallow water as for example drainage ditches (Sitkowska, 1999; Messyasz & Rybak, 2008; Rybak & Messyasz, 2009, 2010; Kowalski, 1975; Mareš, 2009). Shallow aquatic ecosystems are nutrient-rich and high-light environments (Sandgren *et al.*, 2004), on account it status of all of above places indicated high level of nutrient, which stimulate massive macroalgal growth. For the first time, great abundance in Poland of freshwater *Ulva prolifera* O. F. Müller arranged in dense mats (30 m^2) has been reported in small pond near Poznań in 2008 (Rybak & Messyasz, 2009; 2010). Then *Ulva* produced a very large amount of biomass for a short period of time. The reasons of the massive development of green algae from *Ulva* genus in some freshwater ecosystems are not fully investigated.

Expansion of *Ulva* species in inlands water is mainly inhibited by nutrient limitation and a variety of factors. The development of high biomass of *Ulva* thalli is under pressure of herbivores - specially snails, but as in the case of filamentous green algae in good nutrient conditions the grazer control was reduced (Power *et al.*, 2009).

Green algae *Ulva prolifera* O.F. Müller, *Ulva intestinalis* L. and *Ulva flexuosa* subsp. *pilifera* (Kütz.) Bliding occurs in different types of water bodies throughout Poland (Messyasz & Rybak, 2009b). The morphometric characteristic of thalli is acknowledged as a useful feature of *Ulva* when identificated to species. Nutrient contents is one of the factors which influence morphometric features of algae cells and thalli, and also individuality of species, stage of development or features of population.

The main aim of the study was to determine the relationship between uncontrolled presence of *Ulva* thalli mass development in Polish inland water and the content of nutrients in the water ecosystems.

Materials and Methods

The studies were carried out during summer in the years 2009-2011 on the different ecosystems, such as river (Nielba-N), pond (Tulce-T) and reservoir (Malta-M) (Table 1). Sites of the *Ulva* distribution were marked on the map of the Wielkopolska region (Fig. 1). Thalli of *Ulva prolifera* were found in the small pond in the Tulce (village near Poznań), *Ulva intestinalis* in lowland river Nielba (50 km from Poznań) and *Ulva flexuosa* subsp. *pilifera* from Malta reservoir (Poznań). The actual names of the species have been presented by Hayden *et al.* (2003).

The thalli samples of free-floating green algae (Fig. 2a) which were taken from surface by hand were collected into a plastic container and immediately transported to the laboratory. Next, the thalli were repeatedly rinsed with distilled water in order to remove any biotic and abiotic particles attached to them. During each research season about 500 thalli were measured (lengths, widths, presence of proliferations, Fig. 2c). From thalli samples were prepared microscopic slides to observe shape and composition of cells (Fig. 2b) and to measure length and width of cells, number of pyrenoids (staining Lugol's solution) with using a light microscope (40x) and the program ProCap.

The basic physico-chemical parameters of the water (temperature, conductivity, concentration of oxygen and Cl^- as well as the pH level) at the examined sites with *Ulva* were measured with the use of *YSI Professional Plus* handheld multiparameter meter. Water samples were also taken for detailed analyses at laboratory with a view to measuring the content of nitrogen and phosphorus by spectrophotometer HACH DR 2800.

Results and Discusion

Impact of biotic and abiotic factors on the freshwater *Ulva* development.

In general, the massive appearance of these green algae occurred mostly during summer from May or June, with maximum coverage in July or in the beginning of August. Thalli appeared in concentration tightly covering the column of water and were found in two forms - the submerged and free floating all over the surface water. Average concentrations of $N\text{-}NO_3^-$, $N\text{-}NH_4^+$, $P\text{-}PO_4^{3-}$, NaCl and total Fe took out 0.26 $mg.l^{-1}$, 0.86 $mg.l^{-1}$, 0.22 $mg.l^{-1}$, 284 $mg.l^{-1}$ and 0.21 $mg.l^{-1}$ respectively. The value of the electrolytic conductivity was fluctuated around 607 - 1421 $\mu S.cm^{-1}$ (Table 2). Among the studied water ecosystems, the highest values of nitrogen and phosphorus were reported in Tulce pond, where simultaneously were observed dense, monospecific free-floating mats on entire surface.

Table 1. Characteristics of the various *Ulva* patches on research sites in the Wielkopolska region.

Site	Malta Reservoir (M)	Tulce Pond (T)	Nielba River (N)
Location	N 52º24'9.19" E 16º58'13.7"	N 52º20'34.5" E 17º4'39.16"	N 52º48'39.32" E 17º13'18.46"
mean depth (m)	3.1	2.4	0.75
structure of mats	loose	dense	dense
Taxon	*Ulva flexuosa* subsp. *pilifera* (Kützing) M.J.Wynne	*Ulva prolifera* O.F.Müller	*Ulva intestinalis* L.
accompanying	filamentous green algae	macrophytes	filamentous green algae
mats area (m^2)	1-5	30	10-15
Forms	free-floating, submerged	free-floating, submerged	free-floating, submerged
Occurrence	V-VII	V-VIII	V-VII

Table 2. Values (min-max; average) of water main physicochemical factors at the examined sites.

factors / locations	pH	conductivity ($\mu S.cm^{-1}$)	$N\text{-}NO_3$ ($mg.l^{-1}$)	$N\text{-}NH_4$ ($mg.l^{-1}$)	$P\text{-}PO_4$ ($mg.l^{-1}$)	NaCl ($mg.l^{-1}$)	$Fe_{tot.}$ ($mg.l^{-1}$)
Tulce pond	5.03-9.83 (8.3)	655-1421 (978)	0.06-1.11 (0.28)	0.19-4.27 (1.20)	0.02-0.80 (0.44)	61-597 (154)	0.02-0.31 (0.15)
Maltański Reservoir	7.40-8.99 (8.1)	607-805 (714)	0.02-0.14 (0.06)	0.03-1.73 (0.61)	0.01-0.14 (0.05)	98-120 (102)	0.01-0.05 (0.02)
Nielba river	7.40-9.97 (8.6)	719-860 (806)	0.09-0.43 (0.25)	0.22-1.81 (0.54)	0.00-0.23 (0.05)	92-181 (107)	0.01-0.29 (0.05)

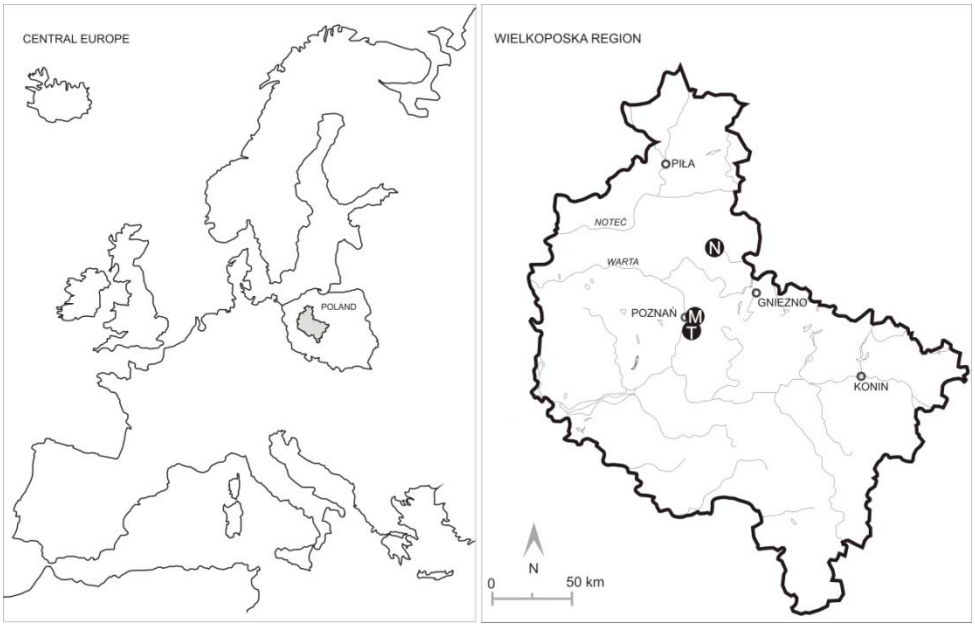


Figure 1. Location of sampling sites for the freshwater *Ulva* in the Wielkopolska region of Poland.
N – the Nielba river; M – the Maltański Reservoir; T – the Tulce pond.

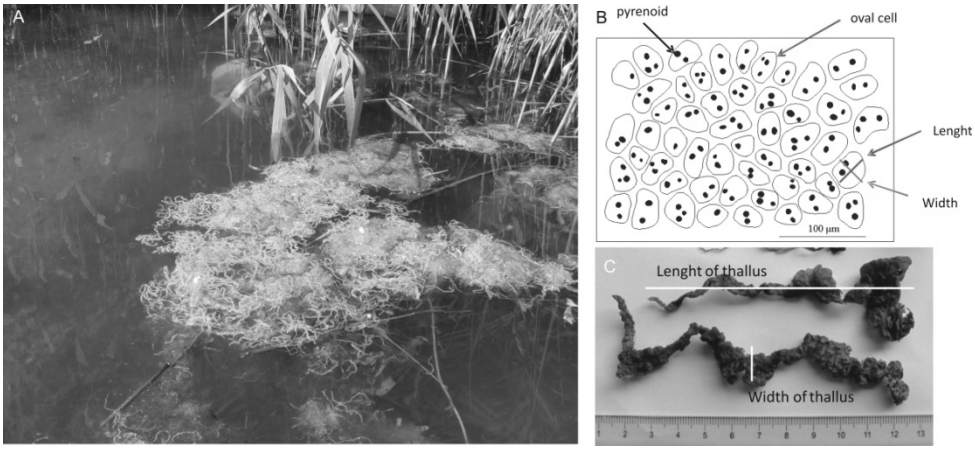


Figure 2. *Ulva* halli in the Maltański Reservoir (A) and the scheme of the measured morphometric features of macroalgal cells (B) and thalli (C).

In addition, thalli achieved average length 20 cm and reached 45 cm maximum and the width from 0.1 up to 4.7 cm (Fig. 3), where thalli collected in May from a pond achieved a much larger size – more than 2 m (Messyasz & Rybak, 2009). The cells had the length from 2.0 up to 24.2 cm and the width in the range from 3.8 up to 17.6 cm (Fig. 4). Length of the thalli suggests that in running waters *Ulva* can reach higher values of these characteristics than in stagnant waters as a result of water flow and habitat conditions. Common situation is with width of the thalli, where in river it is higher than in the reservoir or pond. Therefore, water flow has a significant influence on mats development. For each species (*Ulva intestinali, U. prolifera* and *Ulva flexuosa* subsp. *pilifera*) were observed common results of length and width of thalli and cells, number of pyrenoids (1-3) and shape of cells (oval or square). There were also many differences. Causes of this situation are habitat conditions, e.g. water temperature, nitrogen, phosphorus and, in a lower degree, chloride concentrations. The obtained results demonstrated that even though all water bodies habitats were enriched with N and P they differed from each other in *Ulva* morphology features development of the same species in many respects (size of cells and thalli, young and old thalli participation, presence of branches). Usually, there is a constant number of pirenoids in cells of particular taxon. This phenomenon appeared in *Ulva prolifera* most often. Results of this study highlight the importance of affluence of water in nutrients which allows *Ulva* thalli to obtain good conditions for development shown in its morphometric features.

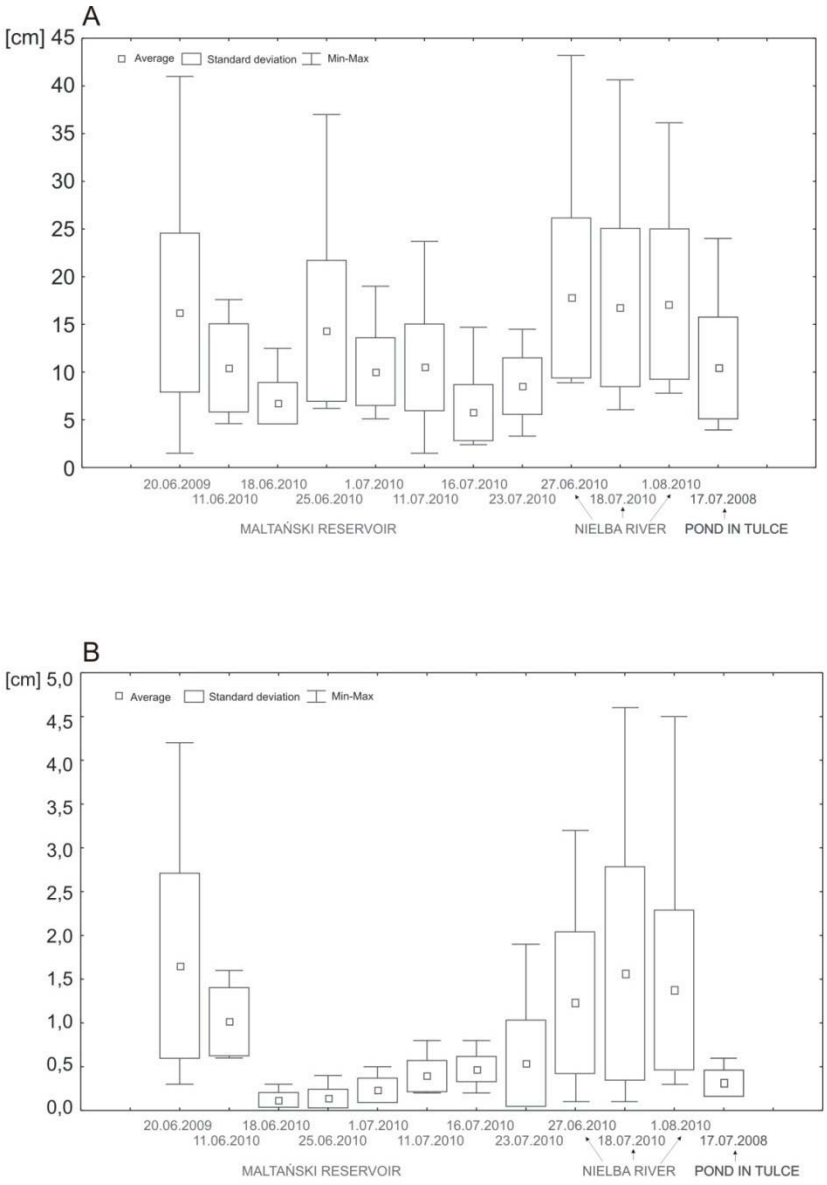


Figure 3. Length (A) and width (B) of *Ulva* thalli in particular types of water body.

The fast growth of thalli in littoral waters, rich in nutrients, was a frequent reason of using *Ulva* biomass in cleaning off waters originating from the fish and shrimp farming (Cohen and Neori, 1991; Neori *et al.*, 1991; Sato, 2006). *Ulva reticulata* thalli implemented into the system of fish breeding ponds grew 4% per

day, removing in this process 6.5 g N m^{-2} day^{-1} (TAN) total ammonia nitrogen from water (Msuya, 2006). It was also made an attempt at composting the biomass of sea species from the *Ulva* genera in order to use it as fertilizer (Wosnitza & Barrantes, 2006). Freshwater *Ulva* which the mass development was observed in the Tulce pond in the period since May by August also influenced the concentration of nitrogen ranked in the water under its thalli. Directly under thalli the concentration of nitrates amounted about 0.20 $mg.l^{-1}$ while, in sites where *Ulva* thalli didn't appear it was higher and gained on average – 0.37 $mg.l^{-1}$ (Fig. 5). We also observed that in individual phases of the freshwater *Ulva* development the ability to remove nitrogen from water was changeable. During appearence of young *Ulva* thalli in the water, under freshwater macroalgal mats concentrations of nitrates amounted from 0.18 to 0.22 $mg.l^{-1}$ at the beginning of the vegetative period. Next, in the course of the thalli development, a decrease in the concentration of nitrates was registered to 0.17 $mg.l^{-1}$ and in the phase of thalli decay repeated increase 0.21 $mg.l^{-1}$ $N\text{-}NO_3$ was recorded. In the end of July an appearance of the second generation of thalli was noted. Young thalli of *Ulva*, which in the sequence of two weeks covered almost entire surfaces of the pond, contributed to the fall in the concentration of nitrates directly under mats to 0.14 $mg.l^{-1}$ (Fig. 6). Under mats with *Ulva*, concentrations of $N\text{-}NO_3$ always were lower than outside them. Observed differences in the $N\text{-}NO_3$ concentration which were noted under and outside mats formed by freshwater *Ulva* were statistically significant (Kruskal-Wallis test H: 5.25; $p< 0.05$). However, in the case of remaining nutrients differences in their concentrations under and outside mats built by freshwater *Ulva* were not statistically significant.

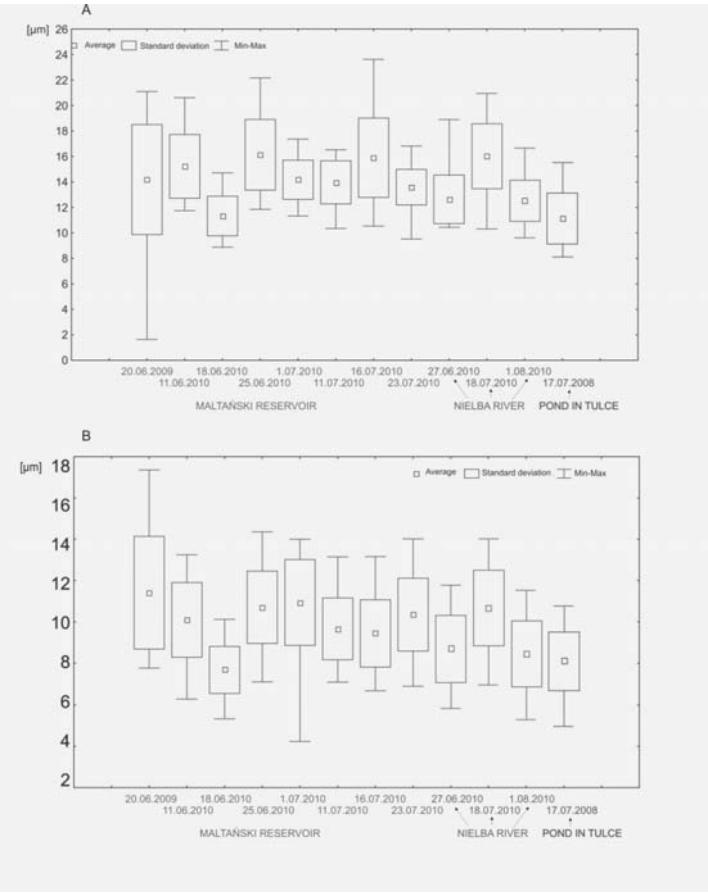


Figure 4. Length (A) and width (B) of *Ulva* cells in particular types of water body.

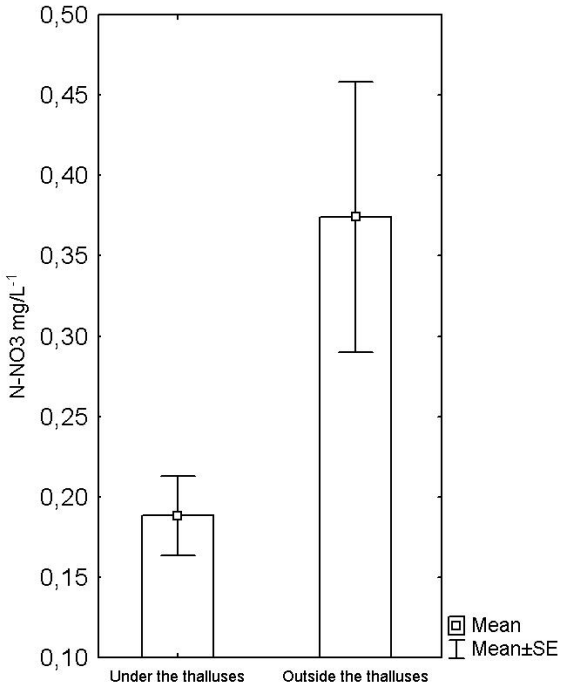


Figure 5. The concentration of nitrate in particular habitats of the pond Tulce.

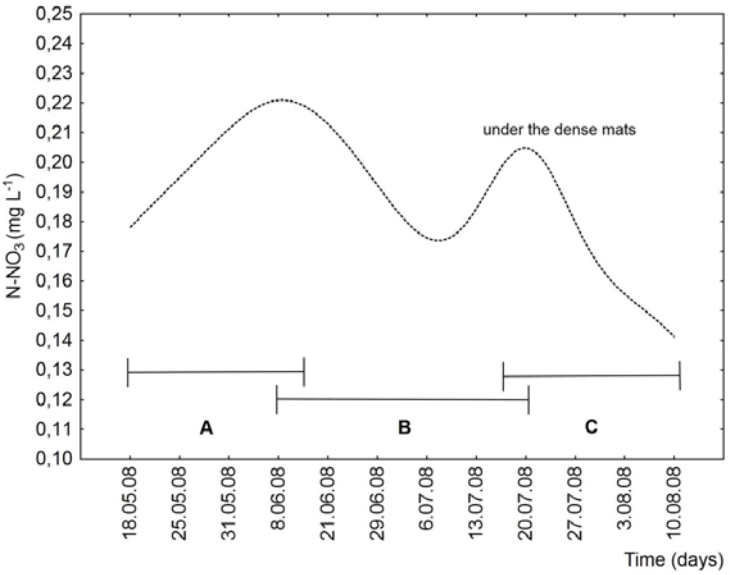


Figure 6. Changes of the concentration of $N\text{-}NO_3$ under the dense mats of *Ulva prolifera* (A – young thallus; B – ripening and decaying thallus; C – young thallus).

Most macrophytes are densely overgrown by periphytic communities (Messyasz & Kuczyńska-Kippen, 2006). Filamentous algae or thalli of macroalgae also constitute an available to the development of attached diatoms. Representatives of the freshwater form of *Ulva* may have two types of thalli - smooth or rough (Messyasz & Rybak, 2010). The first type of *Ulva prolifera* thalli with intestinally-undulating structure was characteristic for young plants while the second type occurs in mature and withering specimens and has a characteristic, curly-bubbled structure. Furthermore, scanning electron microscope observations of *Ulva* thallus revealed that its surface is covered by crystals and on them large numbers of epiphytic diatoms were also noted. During the study period, 89 taxa of diatoms were identified in total. The richest taxonomical structure was recorded for the pond Tulce (63 taxa), the Nielba river (45), while the poorest for the lake Malta (31). The majority of the dominant peryphytic species found on the *Ulva* are cosmopolitan, alkaliphilous, eutrophic and preferring well oxygenated waters. It is interesting to note that species composition did not show much variation in response to thallus growth form. The species *Nitzschia palea* (Kützing) W. Smith, *Cocconeis placentula* Ehr. and *Navicula halophila* (Grunow) Cleve were dominant during all sampling periods. The obtained results revealed that water body type and physical-chemical features of water also seem to play a role in the development of individual differences in epiphytic diatoms assemblages.

Ulva influence for the aqueous ecosystem.

Due to possibility of *Ulva* influence for the aqueous ecosystem, the freshwater thalli consumption was only observed by pond snails (*Lymnaea stagnalis* L.). The freshwater snails observed during natural and experimental conditions were mainly interested in young thalli with a smooth surface and did not generally consume the mature and strongly deformed algae. Freshwater full-grown *Ulva* thalli were characterized by a strongly creased and rough surface. Such structure of a thallus was a result of the mass prevalence of calcium carbonate ($CaCO_3$) crystals and diatom microflora (Messyasz *et al.*, 2010). Consequently, it may be assumed that the occurrence of too developed $CaCO_3$ incrustation may reduce the attractiveness of *Ulva* thalli as a source of food for herbivorous snails. The marine shellfish feeding on periphiton contribute to the increase in light and nutrient accessibility, which enhances the growth of the algal biomass. This type of predation may also control or influence the growth of thalli biomass by other invertebrates eating epiphytic microorganisms (Kamermans *et al.*, 2000). Consequently, it is possible that freshwater species of snails on *Ulva* thalli at freshwater sites (e.g. lakes, ponds and rivers) can consume a large number of periphiton which would stimulate thalli growth.

Moreover, it was found that *Ulva* mats were a very important factor in shaping the phytoseston structure because not all species of algae are tolerating the strongly limited availability of light in their habitats. Only diatoms, cryptophytes and euglenophytes found favourable conditions for their development within *Ulva* mats, irrespective of the site, reaching very high biomass values (on average 4.960 $mg.l^{-1}$ in the pond Tulce; 0.402 $mg.l^{-1}$ in Lake Malta; 0.217 $mg.l^{-1}$ in the Nielba

river). Similar differences were also observed at the level of dominating taxa. The most abundant species in all sites under *Ulva* mats were: *Cryptomonas rostrata* Troitzkaja emend. I. Kiselev (on the average - 18% of the total numbers of alls), *Gomphonema olivaceum* (Horn.) Breb. (15%), *Gomphonema ventricosum* Gregory (12%), *Cocconeis placentula* Ehr. (28-42%), *Gomphonema acuminatum* Ehr. (15%), *Meridion circulare* Ag. (11%), *Cymbella tumidula* Grunow (11%). Microscopic green algae (average biomass 0.098 $mg.l^{-1}$) development in phytoseston communities was strongly restrained by *Ulva* thalli overshadowing. Only in the case of the pond Tulce, green algae achieved much higher biomass than in streams, what was connected with the eutrophic phytoplankton character in all parts of this water body with occurrence of *Scenedesmus*, *Desmodesmus*, *Monoraphidium* and *Tetraëdron*.

Conclusions

The obtained results demonstrated increase in the number of *Ulva* thalli and their size along with the growth of the N and P concentrations in the water. However, the decrease of nutrients concentrations during the *Ulva* development was rather small on account of the permanent supply from the drainage area. The growth of thalli is also affected by a large numbers of epiphytic diatoms (primarily *Cocconeis*) and crystals bands, which excessive contribution may lead to dying of algae.

On the other hand, mass development of these macroscopic algae can contribute to considerable limiting of the light availability in the water, and in consequence reduced the rise in the microscopic algae densities. The fact that *Ulva* thalli are actively consumed by *L. stagnalis*, which used the free floating algal mats as a source of food and shelter, indicates natural character of the *Ulva* phytocoenoses in examined water bodies.

Acknowledgements

The study was supported by the Polish Ministry of Education Science grant No. N N304 0134 37 and partially from projects GDWB-07/2011

References

Apeng, L., Songdong, S., Jianwei, W., Binlun, Y. 2008. Reproduction diversity of *Enteromorpha prolifera*. *Journal of Intergrative Plant Biology*, **50** (5): 622-629.

Bäck, S., Lehvo, A., Blomster, J., 2000. Mass occurrence of unattached *Enteromorpha intestinalis* on the Finnish Baltic Sea coast. *Ann. Bot. Fennici*, **37**: 155-161.

Cohen, I. & Neori, A. 1991. *Ulva lactuca* biofilters for marine fishpond effluents: 1. Ammonia uptake kinetics and nitrogen content. *Bot. Mar*. **34**: 475–482.

Hayden, H. S., Blomster, J., Maggs, C. H., Silva, P., Stanhope, M., Waaland, R. 2003. Linnaeus was right all along: *Ulva* and *Enteromorpha* are not distinct genera. *Eur. J. Phycol*. **38**: 277–294.

Kamermans, P., Malta, E. J., Verschuure, J. M., Schrijvers, L., Lentz, L. F., Tjin, A., Lien , A., 2000. *Grazing by small crustaceans stimulates growth of Ulva ssp. (Chlorophyta) through preferential consumption of epiphytes*. Netherlands Institute of Ecology, Centre for Estuarine and Coastal Ecology, Netherlands.

Kirchhoff, A., Pflugmacher, S., 2002. Comparison of the detoxication capacity of limnic and marine form of the green algae *Enteromorpha compressa*. *Mar. Environ. Res.,* 1–5, 2–73.

Kozłowski, W. 1890. Przyczynek do flory wodorostów okolic Ciechocinka. *Pam. Fizjogr*. **10**, 1-3.

Malta, E., Rijstenbil, J. W., Brouwer, P. E. M., Kromkamp, J. C., 2003. Vertical heterogeneity in physiological characteristics of *Ulva* spp. mats. *Marine Biology*, **143**: 1029-1038.

Mareš, J. 2009. Combined morphological and molecular approach to the assessment of *Ulva* (Chlorophyta; Ulvophyceae) in the Czech Republic. *MSc. Thesis*. Faculty of Science, Univeristy of South Bohemia, Czech Republic.

Marěs, J., Leskinen, E., Sitkowska, M., Skácelová, O., Blomster, J., 2011. True identity of the European freshwater Ulva (Chlorophyta, Ulvophyceae) revealed by a combined molecular and morphological approach. *J. Phycol*, **47.**

Martins, I., Oliveira, J., M., Flindt, M. R., Marques, J. C., 1999. The effect of salinity on the growth rate on the macroalgae Enteromorpha intestinalis (Chlorophyta) in the Mondego estuary (west Portugal). *Acta Oecologia*, **20** (4): 259-265.

Messyasz, B., Kuczyńska-Kippen, N., 2006. Periphytic algal communities: a comparison of *Typha angustifolia* L. and Chara tomentosa L. beds in three shallow lakes (West Poland). *Polish Journal of Ecology*, **54** (1), 15-27.

Messyasz, B., Rybak, A. 2008. Macroalga *Ulva intestinalis* (L.) appearing in freshwater ecosystems of Poland: a new locality in Wielkopolska. *Teka Ochrony i Kształtowania Środowiska Przyrodniczego*, **5**: 126-135 [In Polish].

Messyasz, B., Rybak, A. 2009a. Preliminary morphometic and habitat characteristic of green alga *Ulva prolifera* (Chlorophyta, Ulvophyceae) at freshwater site in the Wielkopolska Region. *Chrońmy Przyrodę Ojczystą,* **65** (5): 347 – 352.

Messyasz, B., Rybak, A., 2009b. The distribution of green algae species from the *Enteromorpha genera* [syn. Ulva; Chlorophyta] in Polish inland waters. *Oceanological and Hydrobiological Studies*, **38** (1): 121-138.

Messyasz, B., Rybak, A., 2010. Abiotic factors affecting the development of *Ulva* sp. (Ulvophyceae; Chlorophyta) in freshwater ecosystems. *Aquat. Ecol*.

Messyasz, B., Rybak, A., Łęska, B., Pikosz, M., 2010. Characteristic of the incrustation covering the thalli of freshwater green alga from the *Ulva* genus (Ulvaceae, Chlorophyta). [Ed:] Schroeder. *Środowisko i Przemysł,* 43-68 [in Polish].

Msuya, F. E., Kyewalyanga, M.S., Dotto, S., 2006. The performance of the seaweed *Ulva reticulata* as a biofilter in a low-tech, low-cost, gravity generated water flow regime in Zanzibar, Tanzania. *Aquaculture,* **254**: 284-292.

Neori, A., Cohen, I. & Gordin, H. 1991. *Ulva lactuca* biofilters for marine fishpond effluents. II. Growth rate, yield and C:N ratio. *Bot. Mar*. **34**: 483–489.

Pliński, M., Jóźwiak, T. 2004. The distribution of water vegetation on the Polish coast of the Baltic Sea in 1996–2000. *Oceanol. Hydrobiol. Stud.* **32** (2): 29–40.

Power, M., Lowe, R., Furey, P., Welter, J., Limm, M., Finlay, J., Bode, C., Chang, S., Goodrich, M., Sculley, J. 2009. Algal mats and insect emergence in rivers under Mediterranean climates: towards photogrammetric surveillance. *Freshwater Biology.*

Rybak, A., Messyasz, B., 2009. The occurrence of macroalgae from the *Ulva* genus (Ulvaceae; Chlorophyta) in Wielkopolska Region. *Bad. Fizjogr. Pol. Zach. Ser. B*, **58**:127-136 [In Polish].

Rybak, A., Messyasz, B., 2010. New locality of greenalga *Ulva prolifera* (Ulvaceae) in Wielkopolska Region. *Fragm. Flor. Geobot. Polonica,* **17** (1): 199-203 [In Polish].

Sandgren, C. D., Engevold, P.M., Neerhof, S., Ehlinger, T. J., 2004. Nuisance Cladophora in Urban Streams: habitats, seasonality, morphology, production, nutrient compositiom, heavy metals, foodweb bottleneck. *In Proceedings of Workshop at the Great Lakes Water Institute*, University of Wisconin-Milwankee, December 8, 2004, 43-56.

Sato K., Ueno Y., Egashira R., 2006. Uptake of Nitrate-Nitrogen in Intensive Shrimp Culture Ponds by Sterile *Ulva* sp. *J. Chem. Eng. Japan*, **39**: 1128–1131.

Sitkowska, M. 1999. Dwa nowe stanowiska *Enteromorpha flexuosa* subsp. *pilifera* (Chlorophyta) w Polsce. *Fragmenta Floristica et Geobotanica, Seria Polonica* **6**, 301–304 [In Polish].

Wosnitza, T., Barrantes J., 2006. Utilization of seaweed Ulva sp. in Paracas Bay (Peru): experimenting with compost. *Journal of Applied Phycology*, **18** (1): 27-31.

Zheng, L., Paul, M. J. 2012. *Effects of Eutrophication on Stream Ecosystems*. Tetra Tech, Inc.

SEASONAL VARIATION OF WATER QUALITY PARAMETERS IN THE CHIRITA RESERVOIR, IASI

IULIANA GABRIELA BREABAN, ANA-MARIA OISTE,
ANDREEA ELENA STOICAN and CATALIN TIBULEAC
Faculty of Geography and Geology, "Alexandru Ioan Cuza" University of Iasi, B-dul Carol I, No. 20 A, 700505, Iasi, Romania, e-mail: iulianab2001@yahoo.com; anamaria.oiste@yahoo.com

Abstract. The study represents the result of a two year monitoring work, with quaterly sampling, from 17 sampling points. The influence of seasonal changes on the physico-chemical characteristics of water resources quality in the Chirita reservoir has been investigated. The study highlights the natural variations of the different water quality parameters of the water lake. Temporal and spatial variations of variables were illustrated through GIS based maps. The pH values of the water samples ranged between 6.856 in winter - to 8.81 in spring, with a mean value for all period of 8.171. The EC and TDS showed the same spatio-temporal patterns, with a range of 539 in autumn and 3240 $mg.l^{-1}$ in spring, with a mean of 969.87 $mg.l^{-1}$, while for phosphates being recorded a range of 0.0203 in autumn - 0.384 $mg.l^{-1}$ in spring with 0.129 $mg.l^{-1}$ mean value. All studied parameters showed significant temporal differences and partial spatial variability. The seasonal changes in water quality were influenced mostly by trophicity, organic pollution, oxide-related process, erosion as well as anthropogenic activities.

Key words: water quality, Chirita reservoir, GIS

Rezumat. Variația sezonieră a parametrilor de calitate a apei din Lacul Chirița, Iași. Studiul reprezintă rezultatul unei activități desfășurate pe parcursul a doi ani de monitorizare, cu prelevare trimestrială de probe, din 17 puncte de pe țărmul lacului. A fost investigată influența schimbărilor sezoniere privind caracteristicile fizico-chimice de calitate a resurselor de apă în lacul Chirița. Studiul evidențiază variațiile naturale ale diferiților parametri de calitate a apei din lac. Variațiile temporale și spațiale au fost ilustrate prin hărți de distribuție realizate cu software GIS. Valorile pH-ului probelor au variat între 6.856 iarna și 8.81 primăvara, cu o valoare medie pe perioada analizată de 8.171. CE si TDS au prezentat același mod de variație spațio – temporală, variind între 539 toamna și 3240 $mg.l^{-1}$ primăvara, cu o valoare medie de 969.87 $mg.l^{-1}$, în timp ce pentru fosfați s-au înregistrat valori între 0.0203 toamna și 0.384 $mg.l^{-1}$ primăvara, cu o valoare medie de 0.129 $mg.l^{-1}$.Toți parametrii studiați au arătat diferențe semnificative în timp, variabilitatea spațială fiind redusă. Schimbările sezoniere ale calității apei au fost influențate în mare parte de fenomene de troficitate, poluare organică, procesele redox, eroziune, precum și de activitățile antropogene.

Cuvinte cheie: calitatea apei, lacul Chirița, GIS

Introduction

This study fits into the general trends of studying water quality and the hydrochemistry of reservoirs of surface waters used as drinking water (Schram, 1971, 1978, Burian, 2002, Grigore *et al.*, 2007, 2009a, 2009b, Scheffer, 2007).

Chirița Lake is located in the North–Eastern part of Iași city, the long side of the lake being oriented from North to South. It was built in 1964 and it has a role of pre-decantation basin of the Prut river water, being an important part of the public water system, ensuring 70% of the distributed water. Moreover, the reservoir was included also into the list of natural reserves due to the role in biodiversity conservation (78 ha of ichthyological fauna reserve). The lake's water quality represents a major concern due to the importance in public health state of the Iasi city inhabitants. During the summer, the eutrophication is one of most important

hydrobiological process, with high potential of deterioration of water quality, so water is directly delivered to the drinking water treatment station Chirita, located in the NE part of the city. The present paper showed some results, which allowed the assessment of water quality for Chirita Lake between 2009 -2011.

Materials and methods

The location of the sampling points was chosen after field observation, the position of pollution sources as well as lake morphology and shape (Fig. 1). Water samples were collected from 17 sampling points (1.5 m from the lake shore and 20-30 cm depth) – quarterly, between September 2009 - June 2011, revealing seasonal variation of some studied parameters.

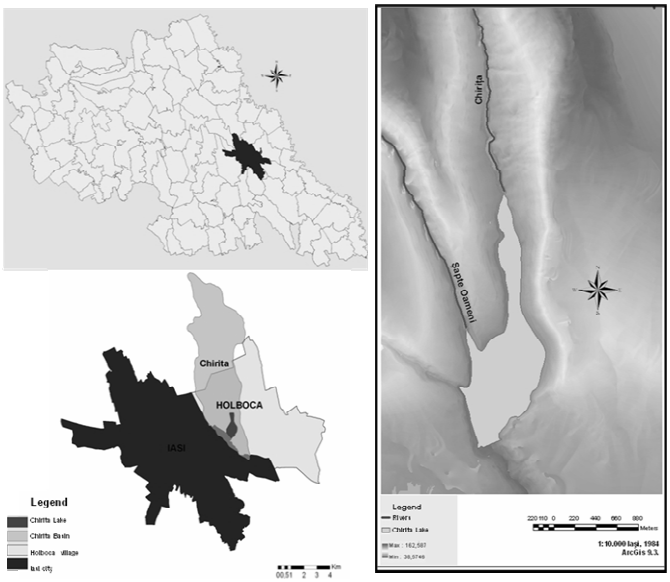


Figure 1. Positioning Chirita lake in Iasi county and landscape in the studied area.

Physical and chemical parameters have been determined in the faculty laboratory: pH, electrical conductivity, turbidity, the oxygen regime (DO, COD-Mn), hardness, alkalinity, chlorides, nutrients - phosphates, nitrates, ammonium, and heavy metals.

The pH, TDS, EC, were determined by potentiometric methods with a Multi 350i/SET WTW multi-parameter instrument, Winkler and the electrometric meter method for oxygen regime, spectrophotometric method using a Shimadzu UV 1601 spectrophotometer for nutrients and atomic absorption spectrometry in air acetylene flames in the hydrochloric solution obtained after the digestion for heavy metals (Trufas, 1975, Evangelou, 1998, Popa, 2001, O'Sullivan, 2004 and Surpateanu, 2007).

The GIS-based mapping using ARC-GIS software 9.3, follow a simple methodology: the lake shape and contour map were digitized, generating the Digital Elevation Model, sampling points coordinates were located with Garmin GPS, data

base with quality parameters joined to attribute table and interpolation of the values for each season using Topo to Raster interpolation method, followed by the reclass. Reclassifying Maps involved the reassignment of the values of an existing map as a function of its initial value of the spatial configuration associated with each map category, keeping the same legend, but representing only the values for each season.

The most relevant variation was highlighted for the phosphates, spatial distribution of the results being correlated with pH and TDS.

Results and discussions

The studied parameters highlight the seasonal variation as well as the spatial distribution within the lake surface. Evaluation of pH, TDS and phosphates may indicate the presence of natural conditions and magnitude of the human impact as well as different quality status of water bodies in analyzed period. The results of the analyzed indicators showed a high variation of the Chirita lake water quality, a strong correlation being established between them.

pH mean values was moderate alkaline 8.17 ± 0.30 for all period with slightly alkaline values only on winter 7.93 ± 0.42. The maxim alkaline value 8.81 was registered in the spring and the neutral value 6.86 in winter, in correlation with atmospheric conditions specific for each season and the regime of the creek that flows into the lake, including the anthropic contribution.

Spatial distribution (Fig. 2) reveal homogeneous distribution of the values during the summer and autumn that are over 8 and the higher variability of the values on winter with neutral values in the lower part of the lake due to the higher depth of the lake and in the spring when that area is located at the confluence area of creek 7 Oameni, an intermittent water course with water flow in this seasons thanks to the snow melting and rainy period.

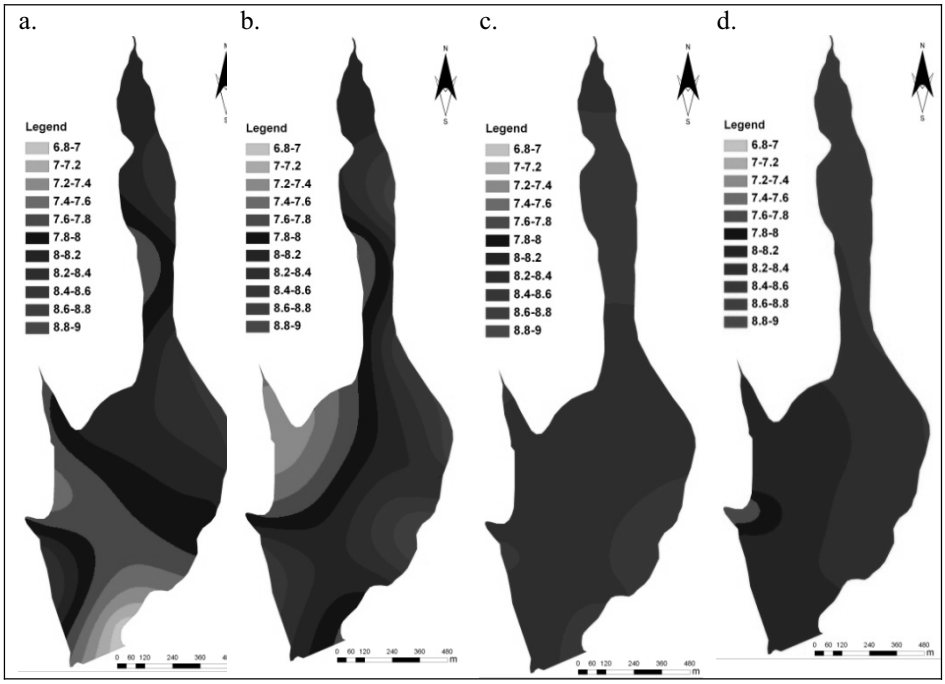


Figure 2. pH distribution map: a. winter, b. spring, c. summer, d. autumn.

TDS is a measure of the combined content of all inorganic and organic substances dissolved in the water, due to the extreme value > 2000 $mg.l^{-1}$, having a negative impact on aquatic ecosystems, moreover in a natural reserve.

TDS registered mean values for all period of 970 ± 321 $mg.l^{-1}$, with higher mean values in the spring 1562 ± 497 $mg.l^{-1}$ without exceeding the > 2000 $mg.l^{-1}$, with the highest value of 3240 $mg.l^{-1}$ in the upper third part of the lake, due to the intake from the Prut river, with a high flow in the spring according to the large catchment and warmer period that cause snow melting, as well as soil erosion on slopes.

Another important aspect is the influence of the 7 Oameni creek that cause higher values. The spatial distribution maps (Fig. 3) reveal homogenous distribution, excepting the spring season with the higher variation on the lake surface.

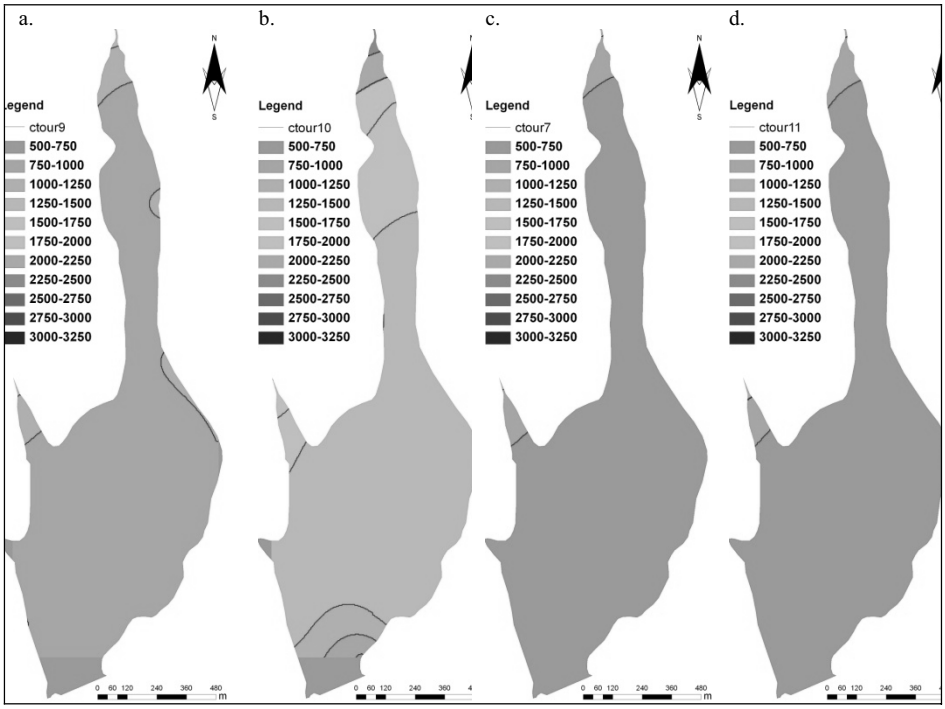


Figure 3. TDS distribution map: a. winter, b. spring, c. summer, d. autumn.

Figure 4. Pollution sources and their effects on Chirita lake water.

A very interesting situation was noticed for phosphates, the most obvious seasonal variation from all parameters analyzed even if are not toxic to people or animals unless they are present in very high levels.

It is a highly reactive, easily measured, water-soluble element and an essential nutrient for plant growth and has a finite reservoir in the natural environment (it does not occur in the atmosphere in gaseous form under ambient conditions) (Macrae *et al.*, 2005, Schippers *et al.*, 2006 and Howden *et al.*, 2009). The sources can be classified as point sources (wastewater effluents) and non point sources (such as applications of lawn fertilizers and disposal of animal wastes), Fig. 4.

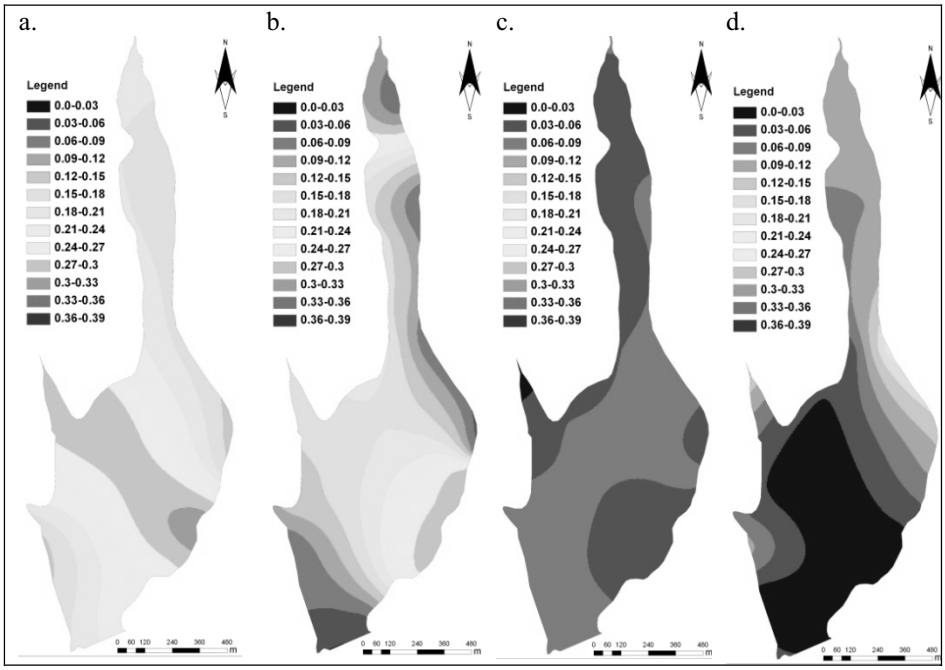


Figure 5. - Phosphates distribution map: a. winter, b. spring, c. summer, d. autumn.

Phosphates (Fig. 5) registered level for all period is 0.129 ± 0.058 mg.l^{-1}, describing a downward trend of mean values from winter 0.210 ± 0.055 mg.l^{-1} that fit in third quality class, to the summer 0.059 ± 0.021 mg.l^{-1} that fit in the first quality class, with the highest value registered in the spring 0.384 mg.l^{-1} without exceeding the limit of third quality class (0.40 mg.l^{-1}). The distribution maps reveal the higher values registered on winter, especially on mid third of the lake due to the larger surface, and an heterogeneous distribution in the spring due to the anthropic influence (Prut river adduction and the large surface of the catchment and predominantly agriculture use of the land) in the upper third of the lake, maintaining the distribution in the mid section.

In the transitional seasons and the summer, the correlation between phosphates and TDS levels becomes obvious, revealing the anthropic influence from

agriculture, known as non point source as well as the eastern shore of the lake used as animal watering area, by the inhabitants.

Conclusions

Water quality is everyone's concern, especially after EU ascension of our country and new legislation implemented concerning water quality, and it is important for all citizens to remember that drinking water quality can be influenced by their actions.

The quality parameters analyzed show correlation between pH, TDS and phosphates levels and the spatial distribution reveal three lake sections with particular characteristic as upper, mid and lower section, supporting different type of influences (natural and anthropic sources).

Acknowledgements

This paper would not have been possible without the help, support, patience, and friendship of some students I had the pleasure to work with. Special thanks to Sorin Victor Rusu, Cosmin Ceucă, Lucian Roşu, Mădălina Paiu, Silviu Şutea – Drăgan.

This work was supported by: Department of Geography, Faculty of Geography and Geology and by the European Social Fund in Romania, under the responsibility of the Managing Authority for the Sectoral Operational Programme for Human Resources Development 2007-2013 [grant POSDRU/CPP 107/DMI 1.5/S/78342].

References

Burian, V. P., 2002. *Lacul de acumulare – supraveghere biologică,* Editura Universităţii de Medicină şi Farmacie, Târgu-Mureş, University Press.

Evangelou, V.P., 1998. *Environmental Soil and Water Chemistry - Principles and Applications*, John Wiley& Sons Inc., New York.

Grigore, P., Nicoară, M. N., Plavan, G., 2009a. Structure of the benthic macroinvertebrates' community from the reservoir Chiriţa – Iasi, in the year 2007, *Studii şi cercetări, Biologie*, Univ. Bacău, **17**: 27–31.

Grigore, P., Nicoară, M. N., Plavan, G., 2009b. The physico – chemical factors influencing the benthic fauna dynamics in the reservoir Chiriţa – Iasi, *Studii şi cercetări, Biologie*, Univ. Bacău, **17**: 68–70.

Grigore, P., Plavan, G., Aoncioaie C., 2007. Aquatic and marshy macrophytes in the ecosystem of Lake Chiriţa Iaşi, *Acta Musei Tutovensis*, Bârlad, **3**: 217–222.

Howden, N., Bowes, M., Clark, A., Humphries, N., Neal, C., 2009. Water quality, nutrients and the European union's Water Framework Directive in a lowland agricultural region: Suffolk, south-east England, *Science of the total environment*, 2966-2979.

Macrae, M. L., Redding, T. E., Creed, I. F., Bell, W. R., Devito, K. J., 2005. Soil, surface water and ground water phosphorous relationships in a partially harvested Boreal Plain aspen catchment, catchment, *Forest ecology and management*. Vol. **206**, no. 1-3.

O'Sullivan, P. E., Reynold, C. S., 2004. *The Lakes Handbook*, Blackwell Publishing, Malden, USA.

Popa, P., Patriche, N., 2001. *Chimia mediului acvatic. Elemente generale de hidrochimie şi ecotoxicologie*, Ed. Ceres, Bucureşti.

Scheffer, M., van Nes, E. H., 2007. Shallow lakes theory revisited: various alternative regimes driven by climate, nutrients, depth and lake size, *Hydrobiologia*, **584**: 455-466.

Schippers, P., van de Weerd, H., de Klein, J., de Jong, B., Scheffer, M., 2006. Impacts of agricultural phosphorous use in catchments on shallow lake water quality: About buffers, time delays and equilibria, *Science of the Total Environment* **369**: 280–294.

Schram, M., 1971. Contribuţii la studiul hidrochimic al lacurilor din Câmpia Moldovei, *Lucrări Ştiinţifice Geografie*, Institutul Pedagogic, Oradea, 146-152.

Schram, M., Erhan, E., 1978. Contribuţii la cunoaşterea potenţialului natural al teritoriului municipiului Iaşi, cu unele implicaţii de ordin practic, A*n. Şt. Univ. „Al.I. Cuza" Iaşi, Secţ. II b*, **XXIV**: 129-131.

Surpăţeanu, M., 2007. *Hidrochimie şi analiza calităţii apei*, Editura Performantica, Iaşi.

Şutea-Drăgan, S., Stoican, A. E., Ţibuleac, C., Rusu, S. V., Breabăn, I. G., 2011. Nutrients regime in the Chirita reservoir, *Lucrări Ştiinţifice Studenţeşti* – vol. **1**/2011.

Trufaş, V., 1975. *Hidrochimie*, Editura Tipografiei Universităţii, Bucureşti.

*** Order no. 161/2006 – *Normative regarding the classification of surface waters in order to establish the ecological status of water bodies.*

*** ArcGIS Desktop 9.3 Help, Environmental Systems Research Institute (ESRI).

NUTRIENTS REGIME VARIATION ON SURFACE WATER IASI CITY URBAN AREA

ANA – MARIA OISTE and IULIANA GABRIELA BREABAN
Faculty of Geography and Geology, Alexandru Ioan Cuza University of Iasi, B-dul Carol I, Nr. 20 A, 700505, Iasi, Romania,Tel: +40 (232) 201480, e-mail: anamaria.oiste@yahoo.com; iulianab2001@yahoo.com

Abstract. Nutrient regime during the year can be correlated with variation caused by season succession, especially temperature changes and other physico-chemical parameters, also by different human activities inside and outside build-up area of Iasi city. Bahlui River is the collector that receives four tributaries (Nicolina, Rediu, Cacaina, Ciric) that cross different functional areas of the city: rural, industrial, residential, with buildings used as collective and individual houses.
Current paper reveal nutrient regime (NH_3, NH_4 şi PO_4) during the dry and wet season (December and June) from 76 sampling points, highlighting temporal and spatial variation between Bahlui river and its tributaries. Field observation followed by lab analyses revealed the variability of nutrients with different sources, natural and anthropic and the quality class they fit: first quality class for NH_4 and PO_4, smaller amounts of NH_3 in the summer and better quality classes for tributaries while a reversed situation for Bahlui River.

Key words: Bahlui River, tributaries, nutrient regime, urban area

Rezumat. Variația regimului nutrienților în apele de suprafață din zona urbană a orașului Iași. Regimul nutrienților este corelat cu variații impuse de succesiunea anotimpurilor, în special de modificarea temperaturii şi variații ale altor parametri fizico-chimici, dar şi de principalele activități din intravilanul si extravilanul oraşului Iaşi. În intravilan, râul Bahlui primeşte patru afluenți (Nicolina, Rediu, Cacaina, Ciric) care traversează zone cu o utilizare variată: rurală, industrială, rezidențială, cu clădiri folosite ca locuințe individuale şi colective.
Această lucrare prezintă regimul nutrienților (NH_3, NH_4 şi PO_4) de-a lungul anotimpurilor rece şi cald (luna decembrie şi iunie) din cele 76 puncte de prelevare, fiind evidente diferențele temporale, cât şi cele spațiale, între râul Bahlui şi afluenții săi. Observațiile în teren, urmate de analiza indicatorilor fizico-chimici, au permis evidențierea variabilității clare a nivelului regimului nutrienților atât din sursele naturale, cât şi cele antropice şi încadrarea lor în diferite clase de calitate: prima clasă de calitate pentru cantitățile NH_4 şi PO_4, NH_3 cu valori mai scăzute în anotimpul cald, încadrându-se în clase de calitate mai bune în cazul afluenților şi situația inversă în cazul râului Bahlui.

Cuvinte cheie: râul Bahlui, afluenți, regimul nutrienților, zona urbană

Introduction

Surface water quality management represent one of the major challenges due to the different pollution sources, being affected by a combination of natural and anthropogenic factors, especially in urban areas, which relative influences change with temporal and spatial scale. Humans influence was highlighted by the changes in river discharges due to urbanization, and discharges from industry, agriculture, or sewerage analyzed during seasonal variation.

Nutrients regime represent the main cause of river area eutrophication that can cause decline in water clarity, increased incidences of algal blooms, taste and odors problems. Ecosystems have been seriously affected by pollution, including wastewater, runoff effluents, land reclamation, recreation and fish culture.

The present study examined the impact of urban areas on nutrient in the surface waters from the build-up area of Iasi city. Were compared average nutrients regime concentrations from a database of water quality and the allowable levels for each parameter and the class it fit, for each section of the analyzed rivers for winter 2010 and summer 2011. The catchments of these rivers ranged from being strongly dominated by urban area to agricultural activities from the outside the city area.

Bahlui River represents the main river that crosses Iasi city with a length, between the spring and the confluence with the River Jijia of 119 km; the monitorized section is about 20 km from the lower part of the catchment. Nicolina River is the main Bahlui river tributary in the south of buildup area of Iasi city; it has a length of 20 km but the monitoring section is about 7.5 km, which means the third lower section of the river basin; for these rivers, monitoring points for water quality exist. From the north part it receives three more tributaries, with different monitoring sections, located in lower section of the river basins: Rediu (1 km), Cacaina (4.1 km) and Ciric (2.1 km) which do not have monitoring points for water quality and flow.

The multi-annual mean discharge is 3.4 $m.s^{-1}$ (Minea, 2006) for Bahlui river and 0.477 $m^3.s^{-1}$, which is determined at hydrometric station in the area Nicolina II district of the city of Iasi (Seliman *et al.*, 2009). Also the climatic data for the city, due to the influence on surface water, are representative: 9.72ºC the multiannual temperature averages at Iasi station 1961-2012 and the 575.5 $mm.m^{-2}$ the multiannual precipitations at Iasi pluviometric point.

Materials and methods

Water samples were collected from 76 sampling points – 42 on Bahlui river and 34 on its tributaries – 15 points for Nicolina river, 7 samples for Rediu and Cacaina and 5 samples for Ciric river in December 2010 and June 2011 revealing seasonal variation (Fig. 1). The location of the sampling points was chosen after field observation and the positions of pollution sources (sewage system locations, dump, drains) and minor riverbed morphology.

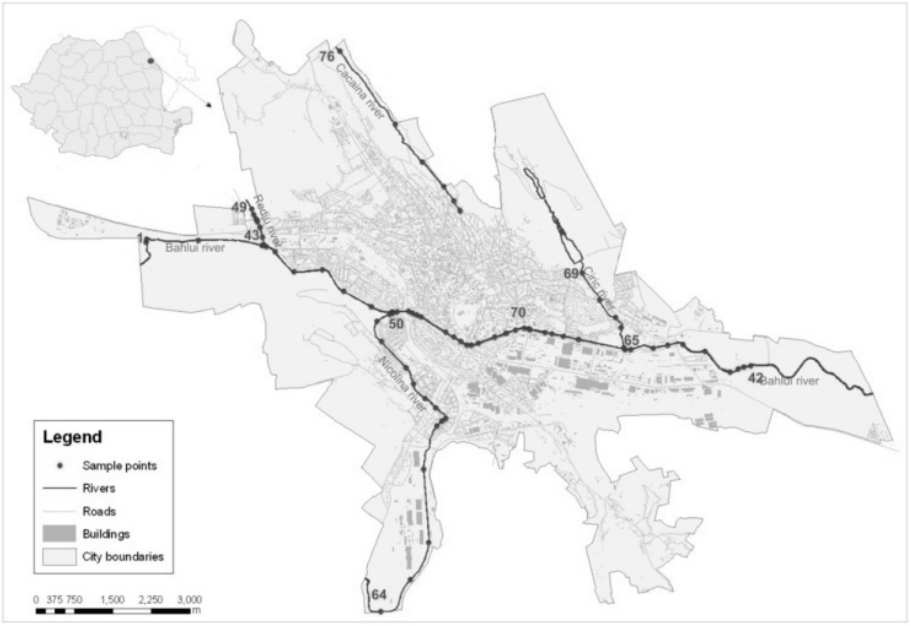


Figure 1. Sampling points location.

More physical and chemical parameters that interact in nutrients regime have been determined: physical and chemical indicators (pH, electrical conductivity, turbidity, the oxygen regime (DO, COD – CCO Mn oxygen consumption and BOD); indicators of the degree of mineralization (chlorides, phosphates, nitrates), physical and chemical selective indicators (hardness, alkalinity), including NH_3, NH_4 and PO_4 concentrations, as well as heavy metals.

The presented parameters are dissolved oxygen - Winckler method was used; for nutrients regime indicators: phosphates - ammonium molybdate reaction, ammonium - Nessler reactive and Seignette salt, nitrates with phenoldisulfonic acid, for spectrophotometric method using a Shimadzu UV 1601 spectrophotometer.

The obtained values were classified according to the Order no. 161/2006, and the class in which the river fits in, revealing seasonal differences and spatial distributions, that representing the Water Framework Directive legislative transposition in Romanian legislation.

Results and discussions

In spite of the importance of nutrients regime indicators, the other physico-chemical parameters as pH, electrical conductivity and dissolved oxygen had an influence in water reactions, being affected by several environmental factors including climate, local biota (plants and animals), bedrock and surface geology, as well as local typology of human activity environmental impacts reflected in surface water quality.

Dissolved oxygen represented the last parameter that was involved in nutrient regime revealing downward trend from winter to summer 11.76 ± 0.18 mg.l^{-1} to 6.30

± 0.12 mg.l^{-1} for Bahlui river and 9.14 ± 0.17 mg.l^{-1} to 3.64 ± 0.17 mg.l^{-1} for tributaries because of the absence of vegetation during the winter (Table 1).

These indicators have an influence also important for biota, the oxygen and the salts dissolved in water (or the EC) and are needed by organisms to survive and as sources of nutrients and energy.

Table 1. Descriptive statistics of the water parameters - dissolved oxygen (DO).

Season	*Statistics*	*Bahlui*	*Rediu*	*Cacaina*	*Ciric*	*Nicolina*
Winter	*Mean*	11.76	1433	2504	8.90	10.22
	Max	12.06	1526	2901	9.05	10.38
	Min	11.50	1296	1680	8.67	10.01
	SD	0.16	119	384	0.16	0.10
Season	*Statistics*	*Bahlui*	*Rediu*	*Cacaina*	*Ciric*	*Nicolina*
Summer	*Mean*	6.30	3.67	3.71	3.59	3.61
	Max	6.50	3.87	3.88	3.87	3.87
	Min	6.01	3.45	3.55	3.25	3.38
	SD	0.12	0.14	0.13	0.25	0.15

The results for the different parameters of nutrients regime are summarized in the box-and-whisker plots of Fig. 2-4. In these plots, the edges of a box are the quartiles, the vertical thick black line is the median, and the vertical thin black lines are the maximum and minimum values, for each season.

Phosphates (Fig. 2) reveal 0.051 ± 0.017 mg.l^{-1} in Bahlui river and 0.048 ± 0.009 mg.l^{-1} in tributaries with higher values on Cacaina river on winter and 0.035 ± 0.015 mg.l^{-1} on Bahlui river and 0.043 ± 0.016 mg.l^{-1} on tributaries in the summer, with higher values in Cacaina river in both seasons: over 0.05 mg.l^{-1} with main sources from agriculture (organic and synthesized fertilizers, and high number of drains on Ticau hill, that represent a source of fecal pollution of surface waters).

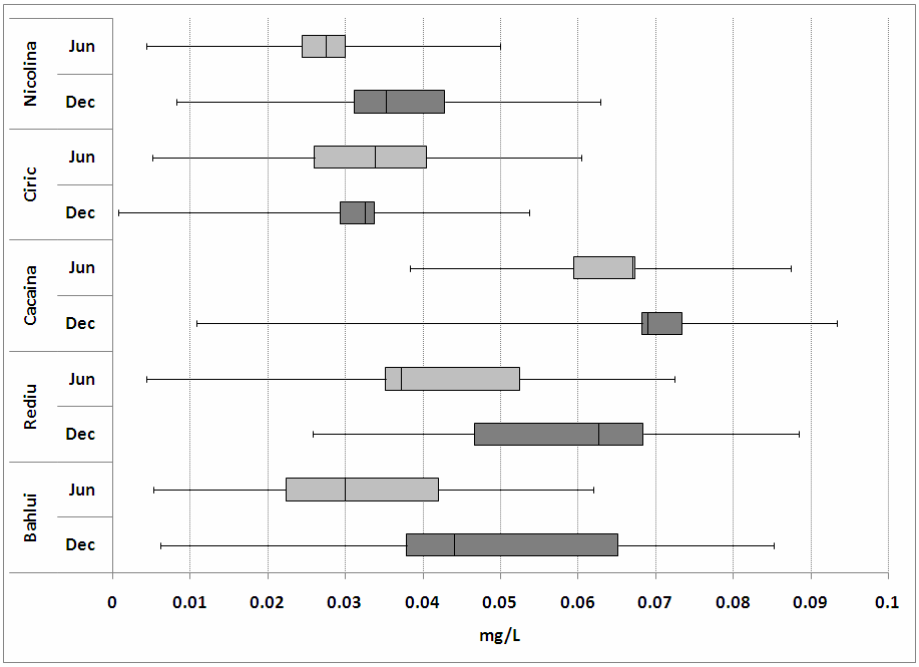


Figure 2. Box-and-whisker diagram for seasonal variation of phosphates.

In the main river were highlighted differences between seasons due to sampling time, during a rainy period, before that phosphates were mobilized by the leakage and can also be correlated with smaller among of dissolved oxygen in the summer, but the values were situated under 0.1 $mg.l^{-1}$ and fit in first quality class for all river sections monitored.

Nitrates (Fig. 3) were derived from three primary sources: rainfall, decomposition of soil organic matter, and nitrogen amendments (fertilizers, manure, etc.), being the last stage of decomposition of organic matter, that indicate an old pollution with the source located far away from the location where was exceeding could be revealed in the river catchment.

The recorded values 2.73 ± 1.26 $mg.l^{-1}$ to 5.73 ± 3.31 $mg.l^{-1}$ for Bahlui river and 16.42 ± 6.59 $mg.l^{-1}$ to 9.99 ± 2.35 $mg.l^{-1}$ for tributaries, with higher values recorded in Cacaina river 33.96 $mg.l^{-1}$ in winter and in Rediu river in the summer 27.67 $mg.l^{-1}$. The values fit in different quality classes showed in Table 2.

According to the different sources, the flow and the weather (rainy or dry period of the sampling) that favored different rate of organic substances decomposition, lower in winter and higher in the summer, the values described a different pattern: an upward trend in Bahlui river, because the higher flow in winter that allow the dilution of the amount of nitrogen, that will be higher in summer due to agricultural activities; the opposite trend is revealed by the tributaries, except the Ciric river that has a number of lakes upstream which supplied with fresh water influencing the nitrogen reaction in water.

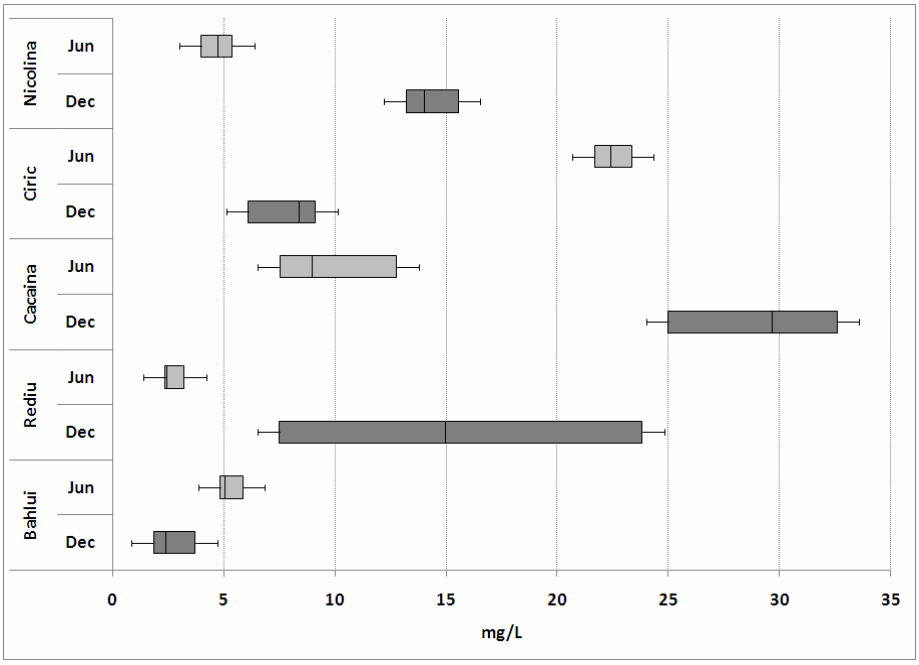


Figure 3. Box-and-whisker diagram for seasonal variation of nitrates.

Ammonium (Fig. 4) indicate a recent pollution of surface waters from different sources as incomplete decomposition of organic substances or from agricultural land, but the registered values in monitored sections fit into first quality class with the limit level 0.4 $mg.l^{-1}$ (Table 3).

In winter, the highest values were registered in Bahlui River 0.036 ± 0.02 $mg.l^{-1}$ with lower values in tributaries, 0.015 ± 0.005 $mg.l^{-1}$, with different evolution in summer 0.027 ± 0.014 $mg.l^{-1}$ in Bahlui River and 0.024 ± 0.021 $mg.l^{-1}$ in tributaries. The differences appear because of different flow: high flow of Bahlui River comparing with the tributaries that show a different rate of organic decomposition, in addition to the presence of more sources of nitrogen as land use in agriculture with fertilizers use. Cacaina River registered higher values in winter due to larger number of drains that flow into the river after crossing an important residential area, and higher values in summer that were registered in Ciric River due to the existence of lakes upstream.

Phosphates represent a limiting nutrient in surface water, in correlation with the amount of available nitrogen (both stages: nitrates and ammonium), causing the cease of plant growth, especially when influxes of nitrogen and phosphorus come from anthropic sources. A high concentration of phosphorus permit plants to assimilate more nitrogen before the phosphorus will be depleted leading to phytoplankton (algae) and macrophyte (aquatic plant) production, higher concentrations of both will support less diversity, causing eutrophication.

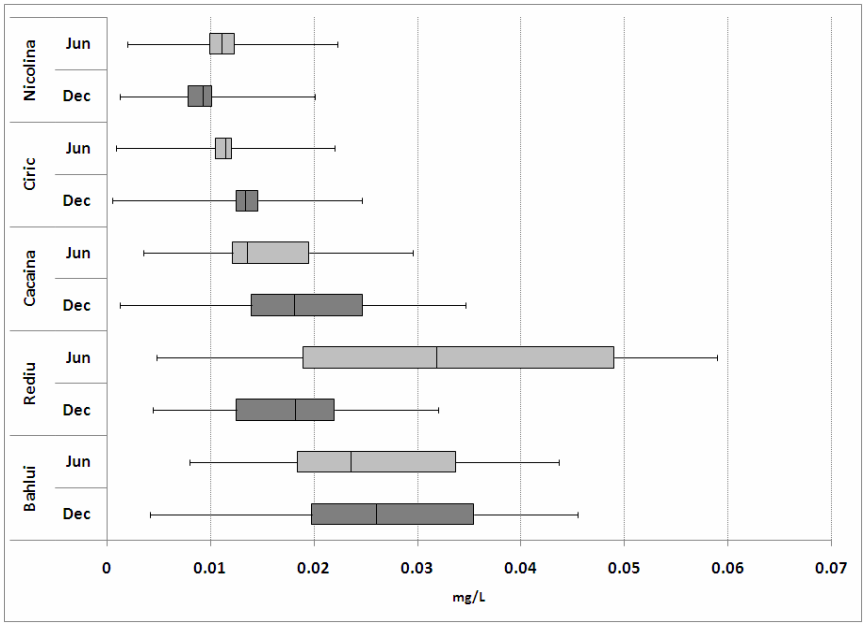


Figure 4. Box-and-whisker diagram for seasonal variation of ammonium.

Table 2. Quality classes from Normative regarding the classification of surface waters in order to establish the ecological status of water bodies.

Parameter	UM	*Class I*	*Class II*	*Class III*	*Class IV*	*Class V*
Phosphates	*mg/L*	*0.1*	*0.2*	*0.4*	*0.9*	*> 0.9*
Nitrates	*mg/L*	*1*	*3*	*5.6*	*11.2*	*> 11.2*
Ammonium	*mg/L*	*0.4*	*0.8*	*1.2*	*3.2*	*>3.2*

Table 3. Quality classes for each section of the analyzed rivers.

River	Season	*Phosphates*	*Nitrates*	*Ammonium*
Bahlui	Winter	Class I	Class II	Class I
	Summer	Class I	Class IV	Class I
Rediu	Winter	Class I	Class V	Class I
	Summer	Class I	Class II	Class I
Cacaina	Winter	Class I	Class V	Class I
	Summer	Class I	Class IV	Class I
Ciric	Winter	Class I	Class IV	Class I
	Summer	Class I	Class V	Class I
Nicolina	Winter	Class I	Class V	Class I
	Summer	Class I	Class III	Class I

Conclusions

The analysis of the nutrients regime indicators and the seasonal variation highlights that:

- From point of view of the quality classes for the nutrients regime, ammonium and phosphates fit into first quality class, only the nitrates concentration represent a real problem, the fifth and fourth quality class being recorded for tributaries, especially in winter due to lower rate of organic decomposition, and the same quality classes in summer for Bahlui and Ciric River.
- The seasonal variations indicates that ammonium and phosphates registered low values in winter and higher in summer, nitrates reveal the reverse situation like, correlated with the variability of decomposition rate of organic substances, and with the pH, dissolved oxygen and electrical conductivity.

Acknowledgements

This work was supported by the European Social Fund in Romania, under the responsibility of the Managing Authority for the Sectoral Operational Programme for Human Resources Development 2007-2013 [grant POSDRU/CPP 107/DMI 1.5/S/78342] and Department of Geography, Faculty of Geography and Geology.

References

Benchea, R., Cretescu, I., Macoveanu, M., 2011. Monitoring of water quality indicators for improving water resources of Bahlui River, *Environmental Engineering and Management Journal*, **10** (3): 327-332.

Cabecinha, E., Lourenc, M., Moura, J. P., Pardal, M. A., Cabral, J. A., 2009. A multi-scale approach to modelling spatial and dynamic ecological patterns for reservoir's water quality management, *Ecological Modelling* 2559–2569.

Chang, H., 2008, Spatial analysis of water quality trends in the Han River basin, South Korea, *Water research*, **42**: 3285–3304.

Chen, D. J., Lu, J., Shen, Y. N, Gong, D. Q., Deng, O. P., 2012. Spatio-temporal variations of nitrogen in an agricultural watershed in eastern China: Catchment export, stream attenuation and discharge, *Environmental Pollution*, **159** (10): 2989-2995.

Cida, F. D., Rosa, A. I., Pardoe, R., Vegae, M., Caviedes-Vidal, E., 2011. Modelling spatial and temporal variations in the water quality of an artificial water reservoir in the semiarid Midwest of Argentina, *Analytica Chimica Acta* **705**: 243– 252.

Crăciun, I., Mitroi, A., Cercel, P., 2004. The Quality Characterization of the Surface Water of the Bahlui Hydrographical Basin, Ovidius University.

Deegan, B. M., White, S. D., Ganf, G. G., 2012. Nutrients and water level fluctuations: A study of three aquatic plants, *River Research and Applications* Volume: **28**, Issue: 3.

Dumitrache, F., Diacu, Diacu, E., 2010. Study on the seasonal variation of nitrogen nutrients content from Ialomita River, *Revista de chimie*, **61** (3): 328-332.

Ekane, N. D., Ebdon, J. E., Huw, D. T., 2011. Integrated analysis of water quality parameters for cost-effective faecal pollution management in river catchments, *Water research*, **45**: 2235–2246.

Gómeza, M. H., Herreraa, S., Soléa, C. D., García-Calvoa, E., Fernández-Albac, A. R., 2012. Spatio-temporal evaluation of organic contaminants and their transformation products along a river basin affected by urban, agricultural and industrial pollution, Science of The Total Environment, Vol. 420, p. 134–145

Hartnett, M., Wilson, J. G., Nash, S., 2011. Irish estuaries: Water quality status and monitoring implications under the water framework directive, *Marine Policy* **35**: 810–818.

Minea, I., 2006. The mean annual discharge in Bahlui Basin, Lucrările Seminarului Geografic "D. Cantemir" nr. **27**.

Minea, I., 2010. The evaluation of the water chemistry and quality for the lakes from the south of the hilly plain of Jijia (Bahlui drainage basin), *Lakes, reservoirs and ponds*, **4** (2): 131-144.

Mourad, D. S. J., van der Perk, M., 2009. Spatio-temporal patterns of nutrient concentrations and export in a north-eastern European lowland catchment, *Hydrological Processes*, **23**, Issue: 13: 1821-1833.

Muntean, A., Coman, M., 2011. The nitrogen regime of the Sasar river, in Baia Mare section, the period 2000-2010, *Aerul şi Apa: Componente ale Mediului*, 495-501.

Neamtu, M., Ciumasu, I., Costica, N., Costica, M., Bobu, M., Nicoara, M., Catrinescu, C., Becker van Slooten, K., Alencastro L., 2009. Chemical, biological, and ecotoxicological assessment of pesticides and persistent organic pollutants in the Bahlui River, Romania, *Environ Sci. Pollut. Res*., (Suppl 1): 76–85.

Oiste, A. M., Breaban, I., G., 2011. Nitrate trends during the season on Bahlui river and its tributaries in Iasi urban area, *Bulletin UASVM Agriculture*, **68** (2): 116–123.

Seliman, C., Plopa, O., Damian, C., Crețu, M., 2009. Urban environment influence on the surface water quality in the City of Iasi, *Lucr. şt., seria Agronomie*, **52**, U.S.A.M.V. Iaşi, 461–466.

*** Order no. 161/2006. *Normative regarding the classification of surface waters in order to establish the ecological.*

THE INFLUENCE OF TALSTAR 10EC INSECTICIDE UPON PHYSIOLOGICAL PARAMETERS IN MARSH FROG (*PELOPHYLAX RIDIBUNDUS*)

ALINA PAUNESCU, GHEORGHITA BRINZEA and MARIA CRISTINA PONEPAL

University of Pitesti, Faculty of Science, Targu din Vale Street, no.1, Pitesti, Romania,
e-mail: alina_paunescu@yahoo.com; georgeta_branzea@yahoo.com

Abstract. The goal of this work is to study the physiological changes (hematological and biochemical indices) induced by the action of Talstar 10EC insecticide in marsh frog (*Pelophylax ridibundus*) at two thermic intervals (4-6ºC and 22-24ºC). Adult male and female frogs were exposed to 0.5 mg biphentrin/g of body weight administrated by intraperitoneal injection, 1 injection at 2 days in a scheme of 3 weeks. We observed increased number of leukocytes, glycaemia value, increased plasma cholesterol and triglyceride, increased hepatosomatic index value and decreased number of erythrocytes. These changes were more powerful in animals treated and kept at 4-6ºC.

Key words: marsh frog, bifenthrin, erythrocytes, leukocytes, glycaemia, cholesterol, triglycerides, hepatosomatic index

Rezumat. Influența insecticidului Talstar 10EC asupra unor indici fiziologici la broasca-de-lac (*Pelophylax ridibundus*). Scopul acestui studiu este investigarea modificărilor unor indici fiziologici (hematologici şi biochimici), determinate de acțiunea insecticidului Talstar 10EC la broasca-de-lac (*Pelophylax ridibundus*) la 2 niveluri termice. Adulții de broască, masculi şi femele au fost supuşi acțiunii a 0.5 mg bifentrin.g^{-1} greutate corporală, administrată prin injecții intraperitoneale, câte 1 injecție la 2 zile, timp de 3 săptămâni. La sfârşitul experimentului am observat o creştere a numărului de leucocite şi a glicemiei, o creştere a nivelului plasmatic de colesterol, trigliceride şi a valorii indicelui hepatosomatic, precum şi o scădere a numărului de eritrocite. Aceste modificări au fost mai pronunțate în cazul animalelor tratate şi ținute la 4-6ºC.

Cuvinte cheie: broasca-de-lac, bifentrin, eritrocite, leucocite, glicemie, colesterol, trigliceride, indice hepatosomatic

Introduction

Populations of amphibians are affected by alterations in their environmental conditions. It is known that insect and snail pests are extensively controlled by using organic pesticides which are a serious environmental hazard (Singhand & Agarwal, 1993). These chemicals may achieve other ecological compartments as lakes and rivers through rains and wind, affecting many other organisms away from the first target (Rand & Petrocelli, 1984).

That why it is necessary to know the effects of these broad spectrum pesticides on aquatic organisms (Elliot, 1977; Casida *et al.*, 1983).

Synthetic pyrethroids came after organochlorines and carbamates and organophosphates became popular not only because of their efficacy but also due to their relatively faster biodegradability and low mammalian toxicity (Khan, 1983).

Pyrethroids belong to the chemical group of nonsystemic insecticides. Talstar 10EC with bifenthrin as active substance ($C_{23}H_{22}ClF_3O_2$ formula) is used for control of insect pest of cotton, vegetables and in public health for control of mosquitoes. Bifenthrin is a type I pyrethroid acting on the central and peripheral

nervous system of invertebrates and vertebrates, namely the channels of Na^+ from nerve endings that closes and opens, resulting in presynaptic membrane depolarization and cell death (Hayes, 1994). It also affects the production of cellular ATP (Roberts & Hutson, 1999).

To determine the effects of disease and unsuitable environmental conditions on amphibians, the normal biochemical and hematological parameters have been investigated. The aim of this study was to investigate the effect of sublethal concentration of bifenthrin on biochemical and haematological parameters of marsh frog *(Pelophylax ridibundus)*.

Materials and methods

In present study we used adult of *Pelophylax ridibundus* of both sexes captured in spring (April-May) from the surrounding areas of the city Piteşti (South Romania). The animals were kept in laboratory condition in aquaterrarios filled with tap water for five days to test their health and accommodate them for the experiment. The water was changed daily to avoid the accumulation of toxic substances.

After 10 days of adaptation in the lab, the frogs were separated in lots, which were separately used for the following experiments: two lots of control individuals, containing animals kept in laboratory at 4-6ºC, respectively at 22-24ºC with no treatment, in running water which was changed everyday, (1) one lot containing animals which were subjected to treatment with Talstar 10EC in a dose of 0.5 $mg.g^{-1}$ of body weight and kept at 4-6ºC, (2) a second lot containing animals which were subjected to treatment with Talstar 10EC in a dose of 0.5 $mg.g^{-1}$ of body weight and kept at 22-24ºC in a thermostatic chamber. Ten animals were used for each lot.

The toxic was administered by intraperitoneal shots, one shot every two days, in a scheme of 3 weeks. The administered dosage of toxic was not lethal as none of the subjects died through the experiment.

At the end of treatment, blood samples were withdrawn from the frogs by cardiac puncture after chloroform anesthesia. The values of operational factors under discussion were determined by using standard automated method: number of erythrocytes and leukocytes was microscopically determined with a Thoma cells numbering chamber, (Picos & Nastasescu, 1988); the glycaemia, cholesterol and triglycerides level has been determinated using an Accutrend GCT.

The hepatosomatic index was calculated using formula:

$$HSI = \frac{liverweight}{bodyweight} \times 100$$

Hematological, biochemical and HIS results were expressed as means ± standard deviation (SD). Statistical analysis was performed as control lot versus treated lot using the Student's t-test. The chosen level of significance is $p<0.05$.

Results and discussions

The action of this pesticide on the liver primarily occurs by increasing the value of hepatosomatic index for the two groups studied (Fig. 1).

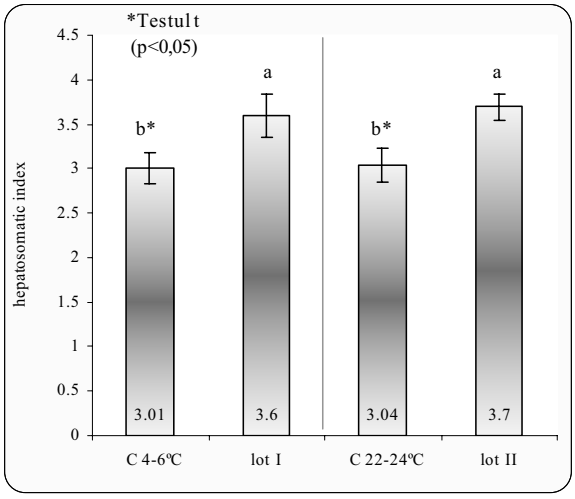


Figure 1. The influence of Talstar 10EC upon hepatosomatic index in marsh frog (b* $p<0.05$ versus control, a $p<0.05$ versus treated lot).

In animals treated with this pesticide and kept at a temperature of 4-6ºC, the index value increased by 19.6% compared to the control value. The same concentration of pesticide caused an increase of the hepatosomatic index value by 25% compared to the control value in animals kept at a temperature of 22-24ºC.

These changes indicates that the liver cells were affected possibly causing an increase in the rate of production of endoplasmic reticulum for the synthesis of protein in liver tissue (Anderson *et al.*, 1998).

The hematological (number of erythrocytes and leukocytes) and biochemical profile (value of glycaemia, cholesterol and triglycerides) provided additional data that have completed the body damage picture induced by the toxic action of Talstar 10EC insecticide on marsh frog (*Pelophylax ridibundus*).

Thus, there is a slight decrease in number of red cells by 7.63%, compared to control value for animals kept at a temperature of 4-6ºC, and 7.19% compared to control value for animals kept at 22-24ºC (Fig. 2).

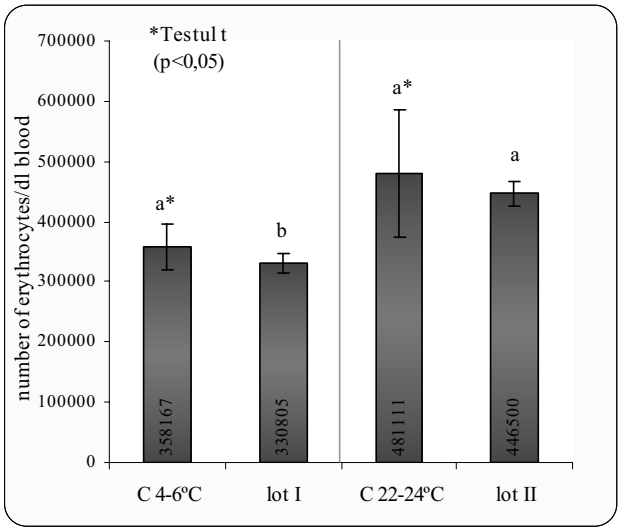


Figure 2. The influence of Talstar 10EC upon number of erythrocytes in marsh frog (b* p<0.05 versus control, a p<0.05 versus treated lot).

Similar changes in the action of Talstar 10EC was observed by Velisek *et al.* (2009) on trout and prussian carp and Ponepal *et al.* (2010) that have studied the effect of this insecticide in three species of fish (prussian carp, perch and bleak). They observed that Talstar 10EC significantly decreased the number of erythrocytes in bleak and perch but had no effect on Prussian carp's erythrocytes.

Talstar 10EC increased number of leukocytes (leukocytosis) in both studied groups (Fig. 3). The analysis of this figure shows an increase by 29.48% in the number of white blood cells compared to control value in animals treated with Roundup®and kept at a temperature of 4-6ºC, while for animals treated with the same concentration of toxic and kept a temperature of 22-24ºC the increase is 23.44% compared to the control value. These changes may have resulted from the excitation of the defense mechanism to counter the effect of the toxicant.

Significant anemia was reported in common carp after acute exposure to delthametrin, a pyrethroid insecticide, but no changes in the blood white cells profile (Svobodova *et al.*, 2003).

Acute poisoning with permethrin decreases the total leukocyte number, especially granulocyte number (Sopinska & Guz, 1998), while acute poisoning with cipermethrin decreases the number of erythrocytes with installation of anemia (Doruncu & Girgin, 2001).

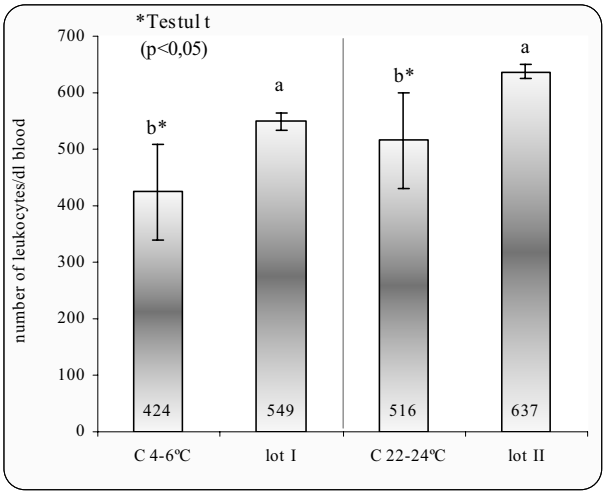


Figure 3. The influence of Talstar 10EC upon number of leukocytes in marsh frog (b* p<0.05 versus control, a p<0.05 versus treated lot).

Biochemical diagnosis provided additional data that have completed the body damage picture induced by this toxic action.

Thus, blood glucose level also changes (Fig. 4) in both studied groups: show an increase by 72.72% compared to control value for animals kept at a temperature of 4-6ºC, and 52.17% compared to control value for animals kept at 22-24ºC. Increased blood glucose level can be explained by reduced tissues utilization of glucose under influence of toxic substance, causing its accumulation in blood.

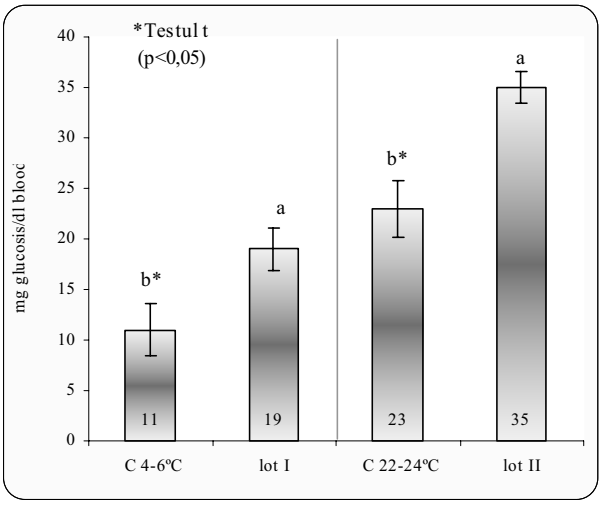


Figure 4. The influence of Talstar 10EC upon glycaemia value in marsh frog (b* p<0.05 versus control, a p<0.05 versus treated lot).

An increase in glucose value was observed in fish (trout and prussian carp) by Velisek *et al.*, (2009a, b) in response to metabolic stress induce by the action of Talstar 10EC insecticide. Jee *et al.*, (2005) also recorded an increase in glucose value and a decrease in quantity of plasma cholesterol and proteins, induced by the action of cipermethrin in *Sebastes schlegeli*.

The toxic substance also works by changing the quantity of plasma cholesterol (Fig. 5) and triglycerides (Fig. 6). Plasma cholesterol records an increased value by 18.99% compared to control group for animals treated and kept at a temperature of 4-6ºC, and 17.14% compared to control for animals kept at a temperature of 22-24ºC.

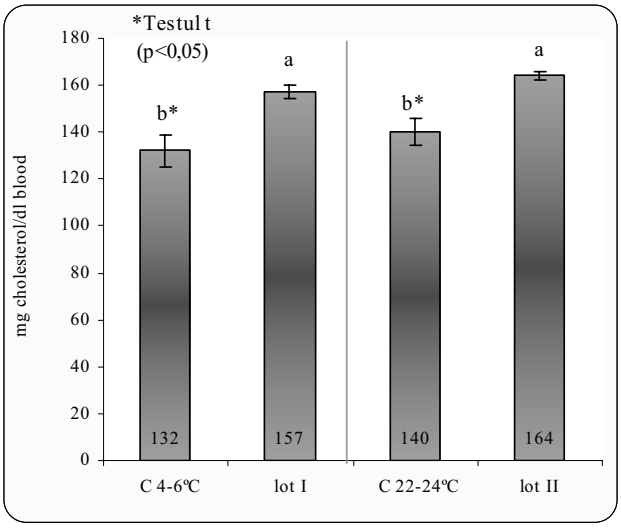


Figure 5. The influence of Talstar 10EC upon cholesterol value in marsh frog (b* p<0.05 versus control, a p<0.05 versus treated lot).

Similar change was observed by Velisek *et al.*, (2006) in trout under the action of cipermethrin.

In terms of triglyceride level (Fig. 6), there was an increase of their value for animals treated and kept at a temperature of 4-6ºC by 11.86% compared to control group, while for animals treated with same concentration of toxic and kept at 22-24ºC this change was not significant.

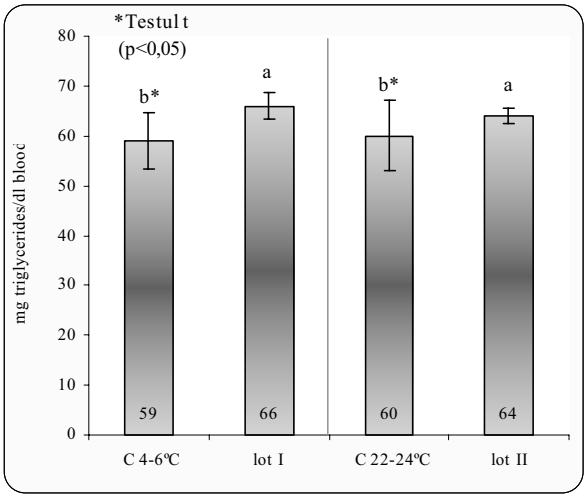


Figure 6. The influence of Talstar 10EC upon triglycerides value in marsh frog (b* $p<0.05$ versus control, a $p<0.05$ versus treated lot).

The action of Talstar 10EC insecticide in concentration of 0.5 $mg.g^{-1}$ body weight upon hematological, biochemical and hepatosomatic index are presented in Table 1.

Table 1. Percentage changes of hematological, biochemical and hepatosomatic index in *Pelophylax ridibundus* upon action of Talstar 10EC insecticide in concentration of 0.5 $mg.g^{-1}$ body weight.

Studied parameters	Time	4-6ºC	22-24ºC
Hepatosomatic index (HIS)	3 weeks	+19.6	+21.71
Number of erythrocytes	3 weeks	-7.63	-7.19
Number of leukocytes	3 weeks	+29.48	+23.44
Glycaemia	3 weeks	+72.72	+52.17
Cholesterol	3 weeks	+18.99	+17.14
Triglycerides	3 weeks	+11.86	+6.66

The analysis of this table shows that Talstar 10EC insecticide is more toxic at low temperature than high temperature. This is probably due to the fact that bifenthrin is more toxic that its metabolites and lower temperature do not favor the metabolism.

The changes of these parameters represent the physyological basis for histological changes induce by the action of Talstar 10EC insecticide.

Conclusions

Hematological and biochemical parameters are suitable tools for assessing environmental influences and stress effects of anthropogenic origin on the condition and health of aquatic vertebrates. After 3 weeks of treatment with sublethal doses of Talstar 10EC insecticide in marsh frog (*Pelophylax ridibundus*) we observed a decreased number of erythrocytes and increased number of leukocytes, glycaemia

value, plasma cholesterol and triglyceride and the hepatosomatic index value. These changes were more powerful in animals that were treated with same concentration of toxic and kept at low temperature.

References

Anderson, T., Forlin, L., Hardig, J., Larsson, A., 1988. Physiological disturbances in fish living in coastal water polluted with bleached kraft mill effluents. *Can. J. Fish Aquat. Sci*., **45**: 1525-1536.

Casida, J. E., Gammon, D. W., Glickman, A. H., Lawrence, L. J., 1983. Mechanisms of selective action of pyrethroid insecticides. *Annual Review of Pharmacology and Toxicology*, **23**: 413–438.

Dorucu, M., Girgin A., 2001. The effects of cypermethrin on some haematological parameters of *Cyprinus carpio*. *Aquaculture International*, **9**: 183–187.

Elliott, M., 1977. Synthetic Pyrethroids. *American Chemical Society*. Washington, 1–28.

Hayes, A. W., 1994. *Principles and methods of toxicology*. Raven Press, New York.

Jee, L. H., Masroor, F., Kang, J.C., 2005. Responses of cypermethrin-induced stress in haematological parameters of Korean rockfish, *Sebastes schlegeli* (Hilgendorf). *Aquaculture Research*, **36**: 898–905.

Khan, N. Y., 1983. An assessment of the hazard of synthetic pyrethroid, insecticides to fish and fish habitat. In: *Pesticide Chemistry; Human Walfare and the Environment* (eds. J. Miyamoto and P.C. Kearney), **3**: 437–450.

Picoş, C. A., Năstăsescu, G., 1988. *Lucrări practice de fiziologie animală*. Tipografia Universității din Bucureşti, Bucureşti, 107, 122-123, 192-195.

Ponepal, M. C., Păunescu, A., Marinescu, A. G., Drăghici, O., 2010. Research on the changes of some physiological parameters in several fish species under the action of the Talstar insecticide. *Analele Universității din Oradea, Fascicula Biologie*, **XVII** (1): 175-179.

Rand, G. M., Petrocelli, S. M., 1984. *Fundamentals of Aquatic Toxicology Methods and Applications*. McGraw-Hill International Book Company.

Roberts, T., Hutson, D., 1999. *Metabolic Pathways of Agrochemicals. Part 2: Insecticides and Fungicides*. The Royal Society of Chemistry, Cambridge, United Kingdom, 1180-1384.

Singhand, A., Agarwal, R. A., 1993. Effect of Cypermethrin on lactate and succinic dehydrogenase and cytochrome oxidase of snail and fish. *Bulletin of Environmental Contamination and Toxicology*,**51**: 445–452.

Sopinska, A., Guz L., 1998. Influence of permethrin on phagocytic activity of carp. *Medycyna Weterynaryjn*, **54**: 126–128.

Svobodova, Z., Luskova, V., Drastichova, J., Svoboda, M., Zlabek, V., 2003. Effect of deltamethrin on haematological indices of common carp (*Cyprinus carpio* L.). *Acta Veterinaria Brno*, **72**: 79–85.

Velisek, J., Wlasow, T., Gomulka, P., Svobodova, Z., Dobsikova, R., Novotny, L., Dudzik, M. 2006. Effects of cypermethrin on rainbow trout (*Oncorhynchus mykiss*). *Veterinarni Medicina*, **51**: 469–476.

Velisek, J., Svobodova, Ć. Z., Machova, Ć. J., 2009a. Effects of bifenthrin on some haematological, biochemical and histopathological parameters of common carp (*Cyprinus carpio* L.). *Fish Physiol Biochem.*, **35**: 583–590.

Velisek, J., Svobodova, Ć. Z., Piackova V., 2009b. Effects of acute exposure to bifenthrin on some haematological, biochemical and histopathological parameters of rainbow trout (*Oncorhynchus mykiss*). *Veterinarni Medicina*, **54** (3): 131–137.

ASSESSING PRESSURE DRIVERS ON BENTHIC MACROINVERTEBRATE AND FISH COMMUNITIES: A CASE STUDY FROM TWO SMALL MEDITERRANEAN RIVERS

THEOCHARIS VAVALIDIS[1,2], DIMITRA C. BOBORI[2] and MARIA LAZARIDOU[1]

[1]Laboratory of Zoology, e-mail: mlazarid@bio.auth.gr

[2]Laboratory of Ichthyology

School of Biology, Aristotle University of Thessaloniki, P.O. Box 134, GR 541 24, Thessaloniki, Greece

e-mail: bobori@bio.auth.gr

Abstract: We examined the impact of anthropogenic pressures on the status of the waters of two small rivers, located in northern Greece, under the view of the Water Framework Directive 2000/60/EC, taking into consideration the communities of benthic macroinvertabrates and fish. Samples of biological elements were taken from 12 sampling stations during one period of low flow and one period of high flow (2010-2011). Additionally, several physical, chemical and hydromorphological parameters, as well as habitat structure modifications were studied. Multivariate statistical analyses were performed (Canoco, Cluster, BioEnv, LinkTree) for extracting the main parameters, driving the structure of benthic macroinvertebrate and fish communities. The impact of anthropogenic pressures and their effect on the ecological quality was investigated by applying the analysis of pressures, based on diffuse and point sources of pollution. In both rivers the average ecological quality, was classified lower than 'good' according to the polymetric STAR_ICMi index and as 'moderate' according to the Hellenic Evaluation System (HES). Total dissolved solids (TDS, $mg.l^{-1}$) and dissolved oxygen (DO, $mg.l^{-1}$) were the most important parameters related to benthic macroinvertebrates, while phosphates (PO_4-P, $mg.l^{-1}$), biological oxygen demand (BOD_5, $mg.l^{-1}$) and substratum, were extracted as the main drivers structuring fish communities. Generally, habitat modification (HMS index) at downstream sampling stations was higher than at those upstream. Furthermore, most of the stations examined exhibited moderate or lower than moderate habitat quality according to the QBR index. The analysis of pressures revealed considerable impact in both river basins. According to the IMPRESS analysis in both rivers the impact of domestic pollution was evident and the risk of not achieving the environmental aims of the Water Framework Directive was evaluated as moderate.

Key words: benthic macroinvertebrates; fish; water quality; Water Framework Directive; IMPRESS analysis

Rezumat. Evaluarea presiunii antropice asupra comunităților de macronevertebrate bentonice și a comunităților piscicole: studiu de caz privind două mici râuri mediteraneene. Am examinat impactul presiunii antropice asupra stării apelor din două râuri mici, situate în nordul Greciei, din punct de vedere a Directivei Cadru Apa 2000/60/CE, luând în considerare comunitățile de macronevertebrate bentonice și pești. Probele biologice au fost prelevate din 12 stații de-a lungul unei perioade cu debit scăzut și a uneia cu debit crescut (2010-2011). În plus au fost studiați câțiva parametric fizici, chimici și hidromorfologici, precum și modificările structurii habitatului. Au fost efectuate analize statistice multivariate (Canoco, Cluster, BioEnv, LinkTree) pentru a obține principalii parametri ce influențează comunitățile de macronevertebrate bentonice și comunitățile piscicole. Impactul presiunii antropice și efectul ei asupra stării ecologice a ecosistemului a fost investigat prin analize privind presiunea antropică, bazată pe sursele de poluare punctuale sau difuze. Calitatea ambelor râuri a fost mai puțin decât "bună" conform indicelui STAR_ICMi şi "moderată" în conformitate cu sistemul de evaluare grecesc Hellenic Evaluation System (HES). Cantitățile totale de solide dizolvate (TDS, $mg.l^{-1}$) și de oxigen dizolvat (DO, $mg.l^{-1}$) au fost cei mai importanți parametri legați de macronevertebratele bentonice, în timp ce fosfații (PO_4-P, $mg.l^{-1}$), consumul biologic de oxigen (CBO_5, $mg.l^{-1}$) și substratul, au fost considerați principalii factori de structurare a comunităților piscicole. În general, modificarea habitatului (indicele HMS), în stațiile de prelevare din aval a fost mai mare decât la cele din amonte. În plus, cele mai multe dintre stațiile examinate au arătat o stare de calitate "moderată" sau mai mică decât "moderată" în conformitate cu

indicele QBR. Analiza presiunii antropice a relevat un impact considerabil în ambele bazine hidrografice. Conform analizei IMPRESS, în ambele râuri impactul poluării a fost evident şi riscul de a nu atinge obiectivele de mediu ale Directivei Cadru Apa a fost evaluat ca fiind moderat.

Cuvinte cheie: macronevertebrate bentonice; peşte; calitatea apei; Directiva Cadru Apa; analiza IMPRESS

Introduction

In European Union, the Water Framework Directive (WFD) 2000/60/EC established a framework for the protection and restoration of waters (inland surface, transitional and coastal waters and groundwaters). The quality classification of waters, according to the WFD is based on biological (phytoplankton, macrophytes and phytobenthos, benthic macroinvertebrates and fishes), hydromorphological and physical-chemical quality elements, with biological elements being of particularly importance.

In the present study we assessed the ecological quality of the surface waters of the rivers Nea Apollonia and Melissourgos (Northern Greece), based on benthic macroinvertebrate and fish communities taking into consideration the anthropogenic pressures.

Materials and methods

Study area

The rivers of Nea Apollonia (catchment area 248.31 km^2, mean altitude 284 m) and Melissourgos (catchment area 194.4 km^2, mean altitude 284 m) are located in Northern Greece, and discharge into the Lake Volvi (Fig. 1). Both rivers present temporary flow regime, with dry periods during the warm months.

Twelve sampling stations (Fig. 1) were selected in the study area, taking into consideration the anthropogenic pressures. Sampling took place during the low flow (LF) period (June –July of 2010), when benthic macroinvertebrates and fish were sampled at 4 stations (A2, A5, M1 and M3; Fig.1), since the rest were dried out. For the high flow (HF) period, sampling took place in all stations during December 2010 for benthic macroinvertebrates and in April 2011 for fish.

Sampling of physical - chemical and hydro-morphological parameters

Water temperature (T, ºC), pH, dissolved oxygen (DO, % and $mg.l^{-1}$), total dissolved solids (TDS, $mg.l^{-1}$) and conductivity (Cond., $\mu S.cm^{-1}$) were measured *in situ* in each sampling station. Additionally two water samples were taken from the surface of the mid – channel for the determination of BOD_5, total suspended solids (TSS, $mg.l^{-1}$) and the concentration of nutrients ($N\text{-}NH_4$, $N\text{-}NO_3$, $N\text{-}NO_2$ and $P\text{-}PO_4$ in $mg.l^{-1}$) according to APHA (1985) methodology.

Substratum was categorized using the six scale particle categories of Wentworth (1922). Water discharge (Disch, $m^3.s^{-1}$) was estimated based on depth and velocity measurements at transect equidistant points of the channel (Horne & Goldman, 1983).

River Habitat Survey (RHS), Habitat Modification Score (HMS) Habitat Quality Assessment (HQA) (Raven *et al.*, 1998) and QBR index (Qualitat del Bosc

Ribera; Munne *et al.*, 2003) were also applied, for assessing the human alterations at each station and for identifying the habitat riparian quality.

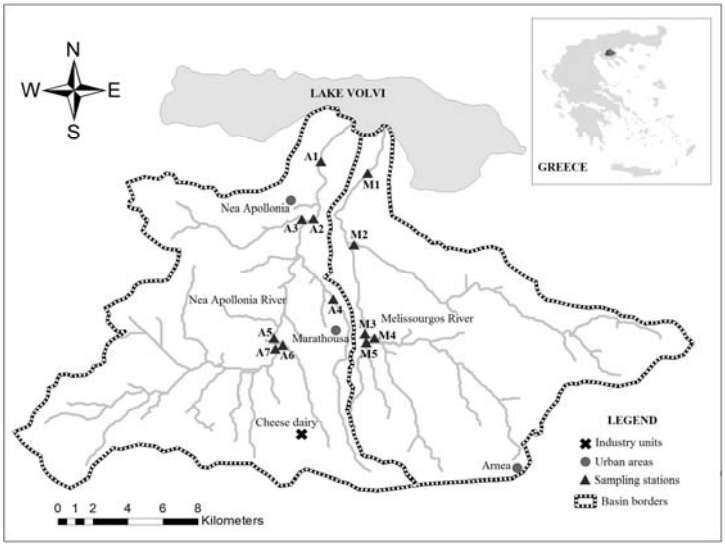


Figure 1. Nea Apollonia and Melissourgos River basins, with the location of sampling stations; point sources of pollution are also indicated.

Sampling of biological elements

Benthic macroinvertebrates were collected with the semi-quantitative 3-minute kick and sweep method (Armitage *et al.*, 1983). The Polymetric STAR_ICMi index and the Hellenic Evaluation System (HES) (Artemiadou & Lazaridou, 2005) were applied in order to assess the ecological quality at the sampling stations.

Fish sampling was carried out by electro-fishing. Fish abundance was expressed as the number of individuals caught per unit of effort, which was set at 100 m^2 of the area sampled (NPUE).

Statistical analyses

Cluster analysis (Primer 6: Clarke & Gorley, 2006) was used to group the sampling stations based on their similarity of benthic macroinvertebrates. Redundancy Analysis (RDA, ter Braak, 1986) was performed with CANOCO (Ter Braak & Smilauer, 1998) to identify possible relationships between benthic macroinvertebrates and environmental variables. Before the RDA, the Monte Carlo permutation test and the inflation factor (<20) were carried in order to select the most significant environmental parameters.

Physical, chemical and morphological data, recorded during the fish sampling, were tested for possible intercorrelations with Spearman's rank correlation analysis; setting the significance of association between the pairs of ranks at |0.9| Furthermore, similarity matrices were created using the Bray Curtis index for fish assemblages and the Euclidean distance for environmental parameters.

Based on these similarity matrices, the BIO-ENV (BEST; Primer 6; Clarke & Gorley, 2006) procedure was applied for extracting the set of environmental variables that was best related to fish assemblages. Moreover, the LINKTREE algorithm was used to optimize the successive binary divisions of fish patterns using threshold values of the environmental parameters extracted with BIO-ENV.

For all analyses data were log (x+1) transformed except for pH and temperature values which were standardized, and dissolved oxygen and substratum (values expressed as percentages) that were arcsine transformed (Zar, 1984).

IMPRESS analysis

Castro *et al.* (2000) methodology was used for assessing the impacts from anthropogenic pressure, evaluating the risk of not achieving the environmental objectives of the Directive and to determine the program of measures to be applied.

Results and discussion

Physical - chemical and hydro-morphological parameters

In both rivers, particularly high values of $N\text{-}NO_2$, exceeding the limits of potable water (JMD Y2/2600/2001) and fish maintenance (Directive 2006/44/EC), were estimated in all samples. Station A6, located after a cheese dairy, and stations A1 and M4, located downstream of the village of Nea Apollonia and the city of Melissourgos respectively, exhibited also high values of $N\text{-}NO_2$, $N\text{-}NH_4$ and $P\text{-}PO_4$, exceeding the above mentioned legislation limits.

According to the Habitat Modification Class, Nea Apollonia River was assessed as obviously modified (average HMS=9), while Melissourgos River as predominantly unmodified (average HMS=7). Riparian habitat quality of Nea Apollonia River (average QBR = 45) and Mellisourgos River (average QBR = 46) was evaluated as poor in both cases. Alterations in the habitat and degradation of the riparian quality occured mainly due to motorways, fluvial terraces and sand extractions.

Biological elements and water quality

A total of 22.550 individuals of benthic macroinvertebrates belonging to 65 different taxa were identified in both rivers. For stations sampled in both periods, the higher abundances occurred during the low flow period. During this period, higher temperatures and less extensive discharge were more favourable for benthic macroinvertebrates maintenance, growth and reproduction (Gasith & Resh, 1999; Lazaridou *et al.*, 2004). In general, A5LF and M3LF stations exhibited the highest macroinvertebrate abundances, while A6HF and M1HF stations the lowest.

Pollution tolerant taxa (e.g. Chironomidae, Baetidae, Oligochaeta) dominated all stations studied in both rivers and in both periods. Station A5HF harbored the highest abundance in moderate to pollution families due to the high Gammaridae presence, while stations A7HF and M5HF had the highest abundances in pollution sensitive families (e.g. Heptageniidae, Philopotamidae, Capnidae).

The two indices applied for assessing the ecological quality at the sampling stations concluded in different results (Table 1), since the HES index includes more taxa in the process for the estimation of water quality. According to

the polymetric STAR_ICMi index, all stations revealed less than good water quality, except station A5LF. According to HES good water quality presented only the stations A5LF, A7HF, M3HF and M5HF while the rest of the sampling stations had moderate and poor water quality.

Table 1. RM types and water quality assessment of the sampling stations, according to the polymetric index STAR_ICMi and the Hellenic Evaluation System (HES) index.

Sampling station	EQR STAR_ICMi	HES
A2LF	0.637	3
A5LF	1.48	4
A1HF	0.27	2
A2HF	0.35	2.5
A3HF	0,52	2
A4HF	0.13	1.5
A5HF	0.13	1.5
A6HF	0.02	2
A7HF	0.66	3.5
M1LF	0.60	3
M3LF	0.57	3
M1HF	0.52	2,5
M2HF	0.61	3
M3HF	0.57	3.5
M4HF	0.59	3
M5HF	0.65	3.5

Fish abundances, (NPUE/100 m^2) are presented in Table 2. All fish were caught in the main reaches of the rivers and no individuals were caught in the tributaries. According to Grossman *et al.* (1998), fish often migrate to the main part of the river where discharge is adequate in order to cope with seasonal dry outs. The higher fish abundances were recorded upstream at low flow period (stations A5LF and M3LF). *Squalius orpheus* was the most abundant species at Nea Apollonia River while *Barbus strumicae* at Melissourgos River (Table 2).

Table 2. Fish abundances (NPUE/100 m^2) per sampling station at Nea Apollonia and Melissourgos Rivers. LF; June, July (2010), HF; April (2010).

Sampling station	*Barbus strumicae*	*Squalius orpheus*	*Cobitis strumicae*	*Alburnus volviticus*
A2LF	0	0	2.5	0
A5LF	7.25	14	0	0
A1HF	0	0	0	0
A2HF	0.5	0	0	0
A3HF	0	0	0	0
A4HF	0	0	0	0
A5HF	0.6	3	0	0
A6HF	0	0	0	0
A7HF	1.5	1.5	0.5	0
M1LF	0.2	5.4	0.6	1.4
M3LF	52.5	13.5	0	0
M1HF	0	1	0.5	0

M2HF	1	0	0	0
M3HF	0.5	0	0	0
M4HF	0	0	0	0
M5HF	0	0	0	0

Statistical analysis

Sampling stations were grouped (CLUSTER analysis) in three different clusters (Fig. 2), based on benthic macroinvertebrate abundances. Clusters a and b were consisted only of Nea Apollonia River stations while all stations of Mellisourgos River were grouped in cluster c. Cluster c was further separated into two sub-clusters, c1 and c2.

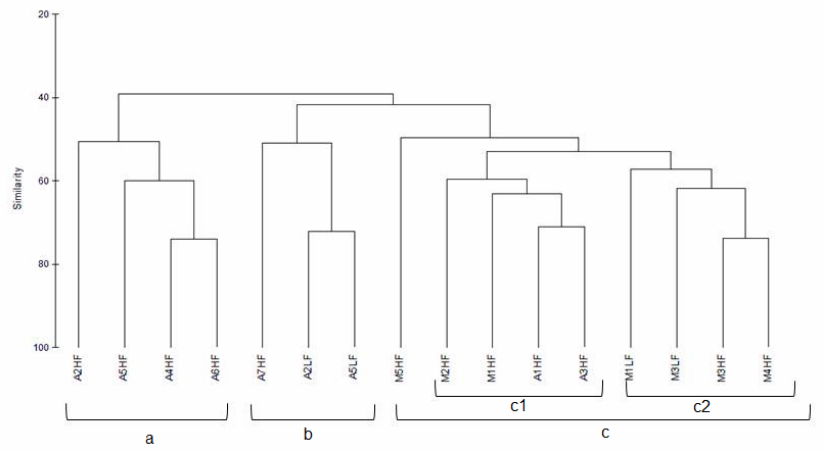


Figure 2. Hierarchical clustering based on benthic macroinvertebrate abundances in Nea Apollonia and Melissourgos River basins.

Dissimilarity between clusters b and c (SIMPER analysis; 56.9%) was mostly due to Gammaridae family (average contribution 12.9%) while the same family was also contributed the most (average contribution 15.8%) to the dissimilarity between clusters b and a (63.9%). Gammaridae, at stations belonging to cluster b is probably related to the cobble substratum recorded at these stations (Bouchard *et al.*, 2004). Moreover, dissimilarity between clusters a and c (55.8%) was mainly due to the taxa Caenidae, Oligochaeta and Baetidae (cummulative contribution 34.7 %). A higher discharge was recorded in Nea Apollonia River (average discharge 1.006 m^3/s) than to Mellisourgos River (average discharge 0.1808 $m^3.s^{-1}$). Caenidae were more abundant in Mellisourgos River, preferring the lower current (Hynes, 1955). Finally, the dissimilarity between the two sub-clusters c1 and c2 (46.6%) was mainly due to the high presence of red Chironomidae in sub cluster c2 (average contribution 12.3 %)). Red Chironomidae is a highly tolerant to pollution taxon (Artemiadou & Lazaridou, 2005), thus stations grouped in c2 cluster exhibit lower water quality that those forming c1 cluster.

According to the Monte Carlo test ($p<0.05$) and the inflation factor (<20) discharge, TDS, DO (%) and N-NO_2 were the only parameters selected for RDA analysis (Fig. 3). The first two ordination axes explained 85.2% of the variance of macroinvertebrate samples. Dissolved oxygen (%) was best correlated with the first axis (0.684) and TDS with the second axis (-0.613). Five out of the seven stations at Melissourgos River had a positive correlation with DO (%), while all seven stations were negatively correlated with TDS ($mg.l^{-1}$). Moreover, all stations at Nea Apollonia River had positively correlated with TDS. The later correlation could be attributed to the high agricultural land coverage at Nea Apollonia River basin (65% of the total area), since intense agricultural activities results at high TDS values (Song *et al.*, 2009). The same was not true for Melissourgos River catchment, were agricultural land cover is up to 33%.

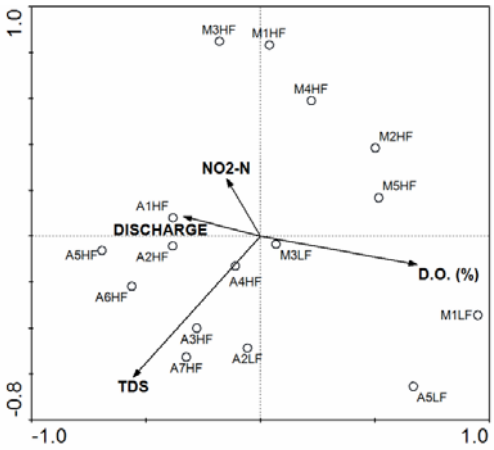


Figure 3. RDA ordination plot of macroinvertebrate samples against physical, chemical and hydro-morphological parameters in Nea Apollonia (A) and Melissourgos (M) Rivers during the low flow (LF) and high flow (HF) periods.

According to Spearman rank correlation, no pair of environmental variables displayed significant correlation ($r<|0.9|$), thus all environmental parameters were used for testing for possible relations with fish abundances.

BIO-ENV analysis extracted ($\rho=0.463$) D.O, $B.O.D._5$, $P\text{-}PO_4$ and substratum (% gravel) as variables, best explaining fish assemblages. Furthermore, the LINKTREE procedure separated the stations where no fish were caught (A, B, C splits; Fig. 4) from the rest of the stations (difference between the two groups B=78%). Moving down the tree graph, split D (B = 46%) separated stations A2LF, M1LF, M1HF from the rest on the basis of $P\text{-}PO_4$ concentration. Further separation of stations (split E, B= 30%; split F, 16%) was based on $B.O.D._5$ while the low B% value ofthe last division (G, B=10%) denoted that stations on these branches were very close and fish abundances did not significantly differ.

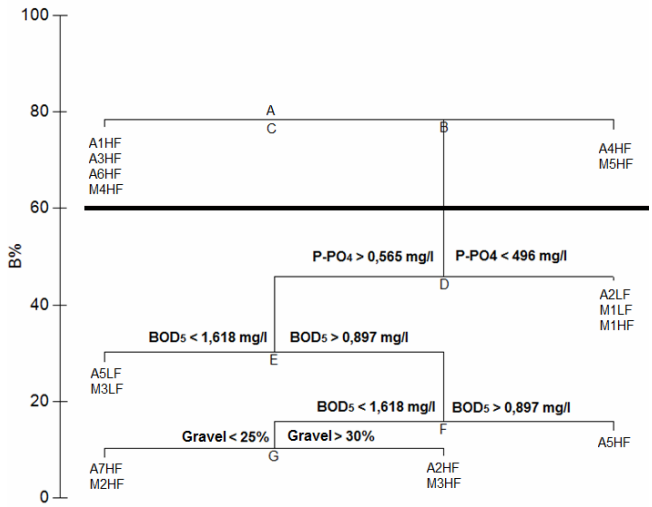


Figure 4. Results of LINKTREE analysis between fish abundances (NPUE) and physical, chemical and hydro-morphological parameters in Nea Apollonia (A) and Melissourgos (M) Rivers during the low flow (LF) and high flow (HF) periods.

Impact and risk analysis

In both river basins, livestock breeding was responsible for the highest inputs of B.O.D.$_5$ while agricultural activities for the total nitrogen and total phosphorus load. In both river basins, the impact of pollution pressures was assessed as 'probable', the probability of failing the environmental objectives of WFD as 'medium' and consequently, a long term program of measures should be applied.

Conclusions

The presence of specific benthic macroinvertebrate taxa, related to water quality, substratum and discharge differentiated the sampling stations. On the other hand, fish were correlated with BOD_5 and phosphates, confirming that different biota differently correspond to the environmental gradient.

The present findings also revealed that considerable pressures occur in both river basins, mainly due to livestock breeding and agricultural activities. Water quality was assessed as inferior to 'good' in most stations, pointing the degradation of the ecological quality in both rivers examined.

Consequently, the failing of the environmental objectives of the WFD is probable in both rivers and thus a long program of measures should be applied.

References

American Public Health Association, 1985. *Standard Methods for the Examination of Water and Wastewater*, 16th Edition, American Public Health Association Inc., Washington D.C.

Armitage, P. D., Moss, D., Wright, J. F. , Furse, M. T., 1983. The performance of a new biological water quality score system based on macroinvertebrates over a wide range of unpolluted running water stations, Water Research, **17**: 333–347.

Artemiadou, V. and Lazaridou, M., 2005. Evaluation Score and Interpretation Index for the ecological quality of running waters in Central and Northern Hellas, *Environmental Monitoring and Assessment*, **110**: 1–40.

Bouchard, R. W., 2004. *Guide to Aquatic Invertebrates of the Upper Midwest, Identification Manual for Students, Citizen Monitors, and Aquatic Resource Professionals*. Water Resources Center, University of Minnesota, St. Paul, MN.

Castro, C. D., Infante A. P., Rodriguez, J. R., , Lerma, R. S. X. and Martinez, F. J. S., 2005. *Manual para la identificacion de las presiones y analisis del impacto en aguas superficiales*, Centro de Publicaciones, Ministerio de Medio Ambiente – Direccion General del Agua.

Clarke, K. R. and Gorely, R. N., 2006. *PRIMER v5. : User Manual/Tutorial*. PRIMER –E, Plymouth.

European Commission, 2000. *Directive 2000/60/EC of the European Parliament and the council 23rd October establishing a framework for community action in the field of water policy*, Official Journal of the European Communities, L327: p. 1–72.

European Commission, 2006. *Directive 2006/44/EC of the European Parliament and of the Council of 6 September 2006 on the quality of fresh waters needing protection or improvement in order to support fish life*, Official Journal of the European Communities, L264: p. 1-12.

Gasith, A. and Resh, V. H., 1999. Stream in Mediterranean Climate Regions: Abiotic Influences and Biotic Responses to Predictable Seasonal Effects, *Annual review of Ecology and Systematics*, Vol. **30**: 51-81.

Grossman, G. D., Ratajczak, R. E., Crawford, M. and Freeman M. C., 1998. Assemblage organization in stream fishes: effects of environmental variation and interspecific interactions, *Ecological Monographs*, **68**: 395–420.

Horne, A. J. and Goldman, C. R., 1983. *Limnology,* McGraw-Hill International Editions, New York.

Hynes, H., 1955. The reproductive cycle of some British freshwater Gammaridae. *Journal of Animal Ecology*, **24**: 352–387.

Joint Ministerial Decision (JDA) Y2/2600/2001, 2001. *Quality of drinking water*, FEK-892/ B'/11-7-01. National Press (Government Newspaper (FEK) and Publications), Athens (in Greek).

Lazaridou, M., Koukoumides, C., Lekka, E. and Gaidagis, G., 2004. Integrative evaluation of the ecological quality of metalliferous streams (Chalkidiki, Macedonia, Hellas), *Environmental Monitoring and Assessment*, **91**: 59–86.

Munne, A., Prat, N., Sola, C., Bonada, N. & Rieradevall, M., 2003. A simple field method for assessing the ecological quality of riparian habitat in rivers and streams: QBR Index. *Aquatic Conservation: Marine and Freshwater Ecosystems*, **13**: 147-163.

Raven, P. J., Holmes, N. T. H., Dawson, F. H. and Everard, M., 1998. Quality assessment using River Habitat Survey data, *Aquatic conservation: Marine and Freshwater Ecosystems*, **8**: 477–499.

Song, M. Y., Leprieur, F., Thomas, A., Lek-Ang, S., Chon, T. S. and Lek, S., 2009. Impact of agricultural land use on aquatic insect assemblages in the Garonne river catchment (SW France), *Aquatic Ecology*, **43** 999–1009.

Ter Brack, C. J., 1986. Canonical correspondence analysis. A new eigenvector technique for multivariate direct gradient analysis, *Ecology*, **67**: 1167–1179.

Wentworth, C. K., 1922. A scale of grade and class terms for clastic sediments, *Journal of Geology,* **30**: 377–392.

Zar, J. H., 1984. *Biostatistical analysis*, Prentice-Hall, Inc., Englewood Cliffs, NJ.

ENVIRONMENTAL ASSESSMENT OF RIVER TROTUS, IN THE TARGU OCNA–ONESTI AREA, BASED ON BENTHIC MACROINVERTEBRATES COMMUNITIES

MIRCEA NICOARA and GABRIEL PLAVAN
„Alexandru Ioan Cuza" University of Iasi, Faculty of Biology, Romania, Bd. Carol I, No. 20 A, 700505,
e-mail: mirmag@uaic.ro, safe2006@yahoo.com

Abstract. The physical – chemical and biological analysis (of macrozoobenthic organisms) of Trotuş River, upstream and downstream of the Targu Ocna town, showed a good water quality status. A possible explanation could be the self purification of water processes, the distance between the first two stations being 5.5 km and 9.8 km from the next two. In addition to the distance between stations, self purification of the water was facilitated by the nature of the substratum (gravel) and water flow high speed.

Key words: Trotus River, benthic macroinvertebrates communities, environmental assessment

Rezumat. Evaluarea ecologică a Râului Trotuş, în zona oraşului Târgu-Ocna, pe baza comunităţilor de macronevertebrate bentonice. În urma efectuării analizelor fizico-chimice şi biologice (macrozoobentos) a râului Trotuş din staţiile din amonte şi aval de oraşul Târgu Ocna s-a evidenţiat o stare de calitate bună a apei. O posibilă explicaţie ar putea fi procesul de autoepurare a apei, distanţa între primele două staţii fiind de 5,5 km, iar între următoarele două de 9,8 km. Pe lângă distanţa mare dintre staţii, autoepurarea a fost facilitată şi de natura substratului (pietros) şi de viteza mare de curgere a apei.

Cuvinte cheie: Râul Trotuş, comunităţi de macronevertebrate bentonice, evaluarea stării de mediu

Introduction

Our studies on the river Trotus aimed at evaluation of the parameters that can characterize the water quality of the river, upstream and down stream of Targu Ocna town. For this purpose we tried to make a biological analysis based on benthic macroinvertebrates communities.

Using physico-chemical aspects, we can elucidate only some questions about the status of water quality (Karr *et al*., 1981; Vega *et al.*, 1998). But, the life forms existing in water have a wide range of reactions against the pollutants or the variations of parameters that characterize the waterbody. It is enough that the values of parameters to undergo a light change to cause a feedback from hydrobionts. This is due to the synergistic effects of substances that are or may occur in the environment at a particular time. Their cumulative effect may be, or may be not harmful to different life forms in the water. After considering the biological perspective, you can verify the nature of substances that influence the hydrobionts in one form or another.

The data obtained as result of the analysis was tried to be reported to the anthropogenic activities, in the areas of sampling, which could influence the hydrobionts.

In the first stage, the stations were chosen for the collection of biological samples. These were placed according to their position in the relation to the settlements along the researched river, respectively towns of Targu Ocna and Onesti.

After fixing the sampling places, biological sampling was carried out. Three samples of the macrozoobenthos and one of water have been collected from each sampling site.

The Trotus Basin has an area of 4349 km^2. Within it, the largest area is represented by mountainous area (66%), Subcarpathians (25%) and the Piemont Trotus – Zabrauti (9%).

Trotus Valley is located between the Tazlău and Casin basins and gives the impression of a long valley, due to its considerable enlargement after leaving the defile from Ciresoaia (Targu Ocna). The maximum enlargement is recorded around Onesti town - a true watershed for Tazlau, Oituz and Casin rivers, towards the landscape descending in wide steps, with the appearance of an amphitheatre (Vacarasu, 1980).

Urban group is formed by six towns: Adjud, Onesti, Targu Ocna, Slanic Moldova, Comanesti and Moinesti. The hypsometric map shows that relief in Trotus area is carried out in steps, the highest being in the upper part of the basin 1400-1600 meters, the lowest in the area of the confluence of the Siret with Trotus River (95-100 meters). The towns are located in valleys, close to water and open places, so explaining the geographical spread on an area of over 75 km between Adjud and Moinesti and a width of 25 kilometres in the central part. By altitude, the towns occupy different positions. The highest is Slanic Moldova (510 meters), the lowest is Adjud (97-105 meters). The others have different positions: Moinesti (440 meters), Comanesti (386 meters), Targu Ocna (257 meters) and Onesti (197-209 meters) (Lupu *et al.*, 1972).

Trotus Valley fragmented the mountains; to the north are the mountains of Tarcau, Gosman and Berzunti, and to the south Ciuc, Nemira and Vranca mountains. The Trotus Valley and other valleys in Bacau County, crossing mountain landscape, present alternations of narrow sectors and expanded sectors.

In the County of Bacau, between the Trotus and Uzu valleys there is the southeast part of the Ciuc Mountains. They are strongly fragmented due to location on the axial zone letting to Trotus Valley, spread from this mountains tentacular spurs, separated by the Sulta and Ciobanasu valleys. The higher altitudes are in the tops of Soiul (1553 meters) and Carunta (1517 meters). These spurs are composed by the Cretaceous-Palaeogene freestone. Structural forms have lost the original character of the North-South oriented heights, thanks to a break through by tributaries on the right of river Trotus (Lupu *et al.*, 1972, Lupu & Văcăraşu, 1980).

Materials and methods

To select the sampling sites we had in mind what areas are upstream and downstream of the two towns found along the river sector (Targu Ocna - Onesti).

We set four sampling sites, one upstream of Targu Ocna (Sampling Site 1), one downstream of Targu Ocna (Sampling Site 2), one upstream the town of Onesti (Sampling Site 3), and the last one situated downstream of Onesti (Sampling Site 4 - Jevreni), in order to be able to see any changes about the diversity of hydrobionts. Three randomly macrozoobenthic samples were sampled from each sampling site using a modified Petersen grab (170.5 cm^2 covered area) and a Surber sampler, and the material was washed through a 0.25 mm sieve.

We preferred to use two types of samplers for a good accuracy of species diversity in Trotus River, taking into account the hetererogeneity of the substratum at each sampling site.

The analysis of the biological material has been carried out in the Hydrobiological Laboratory of the Faculty of Biology of the University "Alexandru Ioan Cuza" of Iasi. The data obtained were statistically interpreted.

Results and Discussion

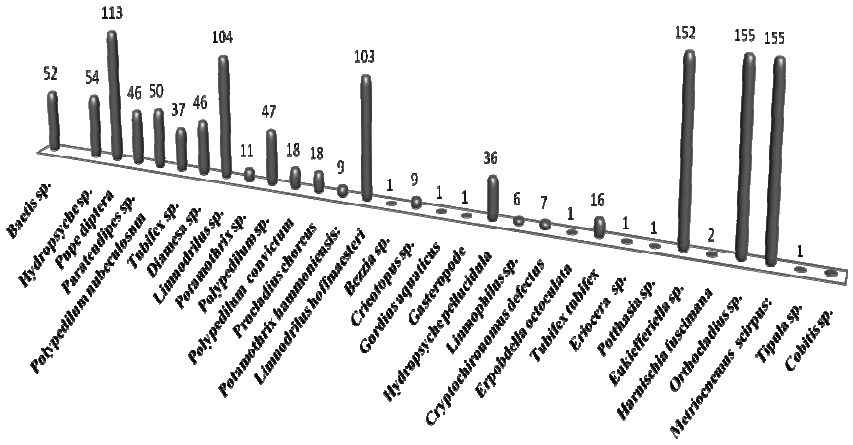


Figure 1. The variance of the numerical abundance of benthic maroinvertebrates in Trotus River at sampling sites I, II, III and IV.

In samples collected from the four sampling sites throughout the sector Targu Ocna (Poeni) - Onesti (Jevreni) 30 species of benthic macroinvertebrates were identified (Fig. 1). The depth level was low in all stations, and the abundance represented by a small number of species. An important abundance had the chironomids species: *Eukiefferiella* spp*., Metriocnemus* ssp., *Orthocladius* spp. and the tubificids: *Limnodrilus* spp. and *Limnodrilus hoffmeisteri*. The species from both families are very cosmopolite, with a high spreading in running waters. *Baetis* ssp., *Hydropsyche* spp*., Polypedilum nubeculosum, Limnodrilus* spp., and *Paratendipes* spp. are relative common here, with low requires regarding the biotope.

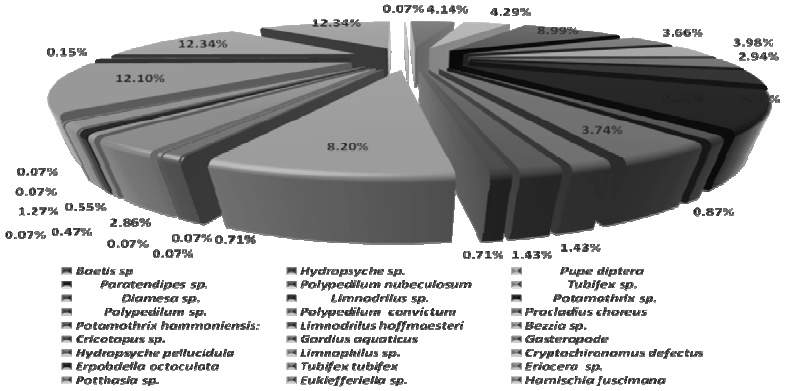


Figure 2. The dominance spectrum of benthic macroinvertebrates in Trotus River at sampling sites I, II, III, IV.

Eukiefferiella ssp., *Metriocnemus* ssp., *Orthocladius* spp. were the dominant species (Fig. 2), participating in a large measure to the production of biomass in the biocoenosis. *Limnodrilus* spp., *Limnodrilus hoffmeisteri* were dominants, *Baetis* spp., *Hydropsyche* spp., *Hydropsyche pellucidula*, *Polypedilum nubeculosum*, *Tubifex* spp., *Diamesa* spp., *Polypedilum* spp., *Paratendipes* spp. were subdominants. Recedent (occasional) species were: *Polypedilum convictum*, *Procladius choreus*, *Tubifex tubifex,* the rest of the species being subrecedents.

The synecological analysis outlined the absence of euconstant species, which were supposed to participate at the fullest extent to achieve the biocoenosis structure and which are supposed to be best adapted to the conditions of the biotope. Two species were identified as constant: *Polypedilum nubeculosum*, *Polypedilum* ssp. (Chironomidae). These are pelofilous species, found in all types of water. In most of the sampling sites, chironomids pupae have been identified, with the same consistency as for the two species mentioned above. Eight species havd the frequency index between 25.1 and 50% (accessory species): *Baetis* spp., *Hydropsyche* spp., *Tubifex* spp., *Potamothrix* spp., *Limnodrilus* spp., *Procladius choreus*, *Limnodrilus hoffmaesteri*, *Tubifex tubifex*, the rest of 19 species being accidental.

At the first sampling site was recorded the lowest number of hydrobiont species (10), but not the smallest number of individuals (220). This may explain the small value of the diversity index. Also the equitability index recorded the lowest value at this station (0.30), the phenomenon being explained by the high abundance of the species *Limnodrilus* spp. (74 individuals of the population registered at the sampling site).

The value of diversity index is influenced by the number of identified species (10) and the value of equitability index (0.31).

Of the four stations analyzed in this sector, the highest value of the diversity index, as well as for the equitability index, was found at the second sampling site ($H_{(s)2}$=3.48; j_2=0.51). At the same time, the number of individuals collected was lower, relative to the other three sampling sites. Although at this

sampling site, the same number of species was found as downstream Onesti (Sampling Site 4), the value of diversity was larger because the individuals are roughly evenly distributed (high equitability).

The fourth sampling site, with a value of $H_{(s)3}$=3.42, had the largest number of species (20) and individuals, comparative to the rest of the sampling sites. However, biodiversity has not the highest value reported to other stations, because the equitability index is low (j_3 = 0.35) and the individuals are not evenly distributed.

The number of species sampled from Jevreni (sampling site 4), corresponds to the number of species in the second station (downstream Targu Ocna), although the index of diversity is lower ($H_{(s)4}$=3.26). The dominance of species *Limnodrilus hoffmeisteri* (68 individuals of 192) explains the difference between the values of the biodiversity at the two stations.

On 25.04.2010, on Trotuş River, Poeni-Jevreni sector, the values of diversity indices were supraunitary at each station: 2.44; 3.48; 3.42 and 3.26 respectively. At the first station we met the lowest value in relation to the other three – 2.44. At the second sampling site was recorded the highest value (3.48), but very close to the value of the third sampling site (3.42).

The high values of the Shannon-Wiener diversity index of benthic macroinvertebrates in Trotus River reveal a specific high diversity within the biocoenosis.

Closest values of the diversity index were found at 2nd and 3rd sampling sites (3.48, respectively 3.42), although the number of individuals found at the second station was the lowest (103) - 12 species, compared to the other stations, and the number of individuals in the third sampling site was the highest (736) - 20 species. This phenomenon can be explained by the high value of the equitability index at the second station (0.51) and the lower equitability index of third station (0.35). So, the low equitability evidence proves strong dominance of the species from the third sampling site: *Eukiefferiella gracei, Orthocladius* spp*., Metriocnemus scirpus*.

The sampling sites 2 and 4 have the same number of species (12), although the values of diversity indices are different (3.48 and 3.26 respectively). Specific diversity of the second sampling site is larger relative to the specific diversity of the forth sampling site, while at the second sampling site there were 105 individuals, at the fourth the total was 192 individuals. Relatively uniform distribution found at station 2 explains this phenomenon, so that the equitability of this station is higher.

Major differences in the values of Shannon-Wiener diversity index were met for the first two stations: at the station 1, $H_{(s)1}$ had the value 2.44, and the second station had $H_{(s)2}$ equal to 3.48. Unlike the second sampling site, where the individuals were relatively evenly distributed, the first sampling site is dominated by *Limnodrilus* spp. (74 of 220 individuals), this bringing with it a lower value of equitability (0.31).

The value of the diversity index is also influenced by the number of species at the two stations (Station 1 with 10 species, and the second station with 12 species).

The first sampling site falls in the Class III of quality - α-mezosaprobic zone - $S_{(st)1}$=2.74. The most of the sapro-indicator species collected from the second

sampling site are characteristic to α-mezosaprobic area (Class II of quality), this sampling site waters having a moderate degree of contamination.

The sampling site from the entrance in the Onesti town has the lowest value of the saprobic index ($S_{(st)3}$=2.11), fitting it into the β-mezosaprobic (Class II of quality). Rocky substratum and speed of the water flow creates a high turbidity, enriching it in oxygen.

The last sampling site had the highest value of the saprobic index ($S_{(st)4}$=2.77). The water in this area is characterized like α-mezosaprobic, with a high degree of impurity. The minor river bed has a thick layer of sediments (50-70 cm).

The lowest value of the saprobic index is at the third sampling site ($S_{(st)3}$ = 2.11). Both the second and third station are belonging to β-mezosaprobic area with moderately polluted waters.

The lowest saprobic index values are recorded at the Poieni station from Targu Ocna's entry and Jevreni sampling site, downstream of the Onesti city. The same sampling sites have smaller values for Shannon diversity index correlated with the other sampling stations. Thus, is emphasized the correlation between the two stations mentioned above. Diversity index values in the second and third stations are very close (3.48 and 3.42). The same stations are included in state II of water quality. High diversity index and state of water quality in these areas, places the two sampling sites (downstream Tg. Ocna and upstream Onesti) above the stations from the upstream and downstream of the studied area.

Conclusions

The area close to the first sampling site (upstream of Targu Ocna) presents the wells for oil extraction and it is situated downstream of Dofteana village (situated at a distance of 500-700 m). On rocky substratum surface from this sampling site there is a film of such oil. This would explain the lower diversity index (2.44) compared to the values of the other three sampling sites. The second sampling site (downstream of Targu Ocna), located at a distance of 5.5 km from the first sampling site, presents the highest diversity richness of the analyzed area ($H_{(s)2}$ = 3.48). This can be explained by the efficiency of self cleaning that takes place in the ecosystem of running water. Diversity index value remains almost unchanged (3.42) at the „upstream Onesti" (sampling site 3).

Near the last sampling site there is located the chemical and petroleum platform of the town Onesti, Jevreni waste water treatment station and land fill of Onesti town. However, the diversity index is not significantly changed compared to previous sampling site, $H_{(s)4}$=3.26. The decreasing value of this index can be explained by the presence of three species eudominant: *Eukiefferiella* spp., *Metriocnemus* spp., and *Orthocladius* spp.

As we have seen in the synecological analysis and the Shannon diversity index, the first sampling site (upstream Targu Ocna) is characterized by a low diversity and equitability of macrozoobenthos, but also by a strongly contaminated water, given the high value of saprobic index ($S_{(st)1}$=2.74). Both oil extraction wells and Dofteana and Poeni villages' neighborhood can be pollution sources and causes of the water state that characterize the study area.

Downstream Targu Ocna (second sampling site), showed the highest macrozoobentos diversity, very close to the third sampling site, and falls in the moderate contamination ($S_{(st)2}$=2.47) cathegory. The lowest value of the saprobic index was encountered at the third sampling site (upstream Onesti), so the area is characterized by a low water contamination ($S_{(st)3}$=2.11). This low values is due the self purificaton of river between the first and second sampling sites, by a distance of 5.5 km and between 2nd and 3rd sampling site, where the distance is about 9 km.

Jevreni sampling site (number 4- downstream Onesti), located in the vicinity of petrochemical platform, the town landfill and wastewater treatment station, presents the highest value of saprobic index ($S_{(st)4}$=2.77). The area close to the fourth sampling site has a strong water contamination (α-mezosaprobic area), with many species of oligochaete and a lower level of dissolved oxygen compared to the other stations. Flow is slow, muddy substratum, unable to achieve highly oxygenated water in the area.

References

Karr, R. J. and Dudley, R. D., 1981. Ecological perspective on water quality goals, *Environmental Management*, **5**, Issue 1: 55-68.

Lupu, N., Văcăraşu, I., 1980. *Tipuri de aşezări in bazinul superior al Trotuşului*, p. 67-71, Ed. Ştiinţifică Bucureşti.

Lupu, N., Văcăraşu, I., Branduş, C., 1972. *Judeţele patriei-Judeţul Bacău*, p. 27-30, Ed. Ştiinţifică Bucureşti.

Văcăraşu, I., 1980. *Valea Trotuşului: monografie*, Bucureşti: Sport-Turism.

Vega, M., Pardo, R., Barrado, E. and Deban, L., 1998. Assessment of seasonal and polluting effects on the quality of river water by exploratory data analysis, *Water Research*, **32**, Issue 12: 3581–3592.

RESEARCHES ON THE FISH COMMUNITIES IN THE MIDDLE BASIN OF RIVER MURES IN 2009-2011

DOREL URECHE[1], RAMONA PINTILIEASA[2], CAMELIA URECHE[1] and ROXANA ELENA VOICU[1]

[1] University „Vasile Alecsandri" of Bacau, Marasesti street, no. 157, 600115, Bacau, Romania, e-mail: dureche@ub.ro; urechec@ub.ro;

[2] „Alexandru Ioan Cuza" University of Iasi, Bd. Carol I, 20A, Iasi, Romania, e-mail: bio06ramona@yahoo.com

Abstract. The study was carried out in the middle basin of River Mures, on the main course of the river and on some of its tributaries over the period 2009-2011. Some of the physico-geographical and hydrochemical parameters were determined. The aim of the study was to assess the actual state of fish communities in the middle basin of River Mures, and also to highlight significant changes in fish communities, reflected by the level of biodiversity index, numerical stock, weight stock, and biological integrity. The biological material was collected by electrofishing from 21 sampling sites. Over the period 2009-2011, 27 fish species were found, with an amount of 5,649 individuals and 33,214.6 g. One of the 27 fish species is a non-native species (*Pseudorasbora parva*) and 26 are native species. The numerical stock at sampling sites ranged between 21.19 and 502.2 ind.100 m^{-2} while the weight stock has recorded values ranging between 136.72 and 3,760.06 g.100 m^{-2}. Based on the ecological indices, three fish zones were found in the basin of the River Tarnava Mica: brown trout zone, mediterranean barbel zone and chub zone. Unlike the situation recorded in the past, the fish zones are changed. Thus, the upper part of the common nase zone was replaced by the mediterranean barbel zone while the barbel zone and the lower part of common nase zone were replaced by the chub zone.

Key words: fish communities, biological integrity, River Mures

Rezumat. Cercetări asupra comunităților piscicole din bazinul mijlociu al râului Mureș în perioada 2009-2011. Studiul s-a desfășurat în perioada 2009-2011, pe cursul principal al râului Mureș și pe unii afluenți. Au fost determinați unii parametri fizico-geografici și hidrochimici. Scopul studiului a fost de a estima starea actuală a comunităților de pești din bazinul mijlociu al râului Mureș și de a evidenția modificările semnificative ale acestora, reflectate de nivelul indicelui de biodiversitate, stoc numeric și gravimetric, și integritate biologică. Au fost identificate 27 specii de pești cu un total de 5649 indivizi și o biomasă de 33214,6 g. Una din cele 27 de specii este non-nativă (*Pseudorasbora parva*) iar 26 sunt native. Stocul numeric a avut valori cuprinse între 21,19 și 502,2 ind.100 m^{-2}, iar cel gravimetric între 136,72 și 3760,06 g.100 m^{-2}. Pe baza indicilor ecologici, în bazinul mijlociu al râului Mureș au fost identificate trei zone piscicole: zona păstrăvului, zona moioagei și zona cleanului. Spre deosebire de situația din trecut, zonele piscicole sunt modificate, astfel că porțiunea superioară a zonei scobarului a fost înlocuită de zona moioagei iar porțiunea inferioară a zonei scobarului și zona mrenei au fost înlocuite de zona cleanului.

Cuvinte cheie: comunități piscicole, integritate biologică, Râul Mures

Introduction

The study was carried out in the middle basin of the River Mures, on the main course of the river and on some of its tributaries over the period 2009-2011. The aim of the study was to assess the actual state of fish communities in the middle basin of River Mures, and also to highlight significant changes in fish communities.

The middle basin of River Mures is located in the central part of Transilvanian plateau, between Deda and Alba Iulia, and includes about one third of the total lenght of the river together with the corresponding tributaries. Aries,

Tarnavele and Sebes Rivers are the main tributaries in the middle basin of River Mures. The boundaries of investigated area are Mures downstream Sebes and Lechinta, upstream Lechinta.

Material and methods

The biological material was collected by electrofishing from 21 sampling sites on the main course of the River Mures and on some of its tributaries (Sebes, Paraul Carbunarilor, Fitcau, Ripa, Idicel, Nadasa, Beica, Lutul, Niraj, Nirajul Mare and Lechinta). All of the fish individuals were determined and biometrical processed.

Qualitative and quantitative methods were used, in order to assess the overall state of fish communities. Based on the ecological indices, the structure of fish communities at sampling sites was set, as well as the fish zones of the middle basin of River Mures. The assessment of aquatic environment quality was possible using the biodiversity index, and the biological integrity index (IBI).

Results and discutions

Over the period 2009-2011, 27 fish species were found, with an amount of 5,256 individuals and 32,438.6 g. One of the 27 fish species is a non-native and 26 are native species.

It was noticed the lack of a number of 11 species of the 40 found by Banarescu in the area where the study was carried out in 2009-2011. On the other hand, over the period 2009-2011, four new fish species were found in addition to the situation recorded by Banarescu in 1964, respectively *Leuciscus idus, Pseudorasbora parva, Carassius gibelio,* and *Sabanejewia balcanica*.

Changes in the composition of fish communities are due to the extension of the spreading area of some of the most resistant species. In the same time, some of the most sensitive fish species have special ecological requirements and a more limited area of distribution.

The number of fish species at sampling sites varied between 1 (Nirajul Mare, upstream Campul Cetatii) and 15 (Lechinta, upstream Lechinta) (Fig. 2).

Numerical stock

The numerical stock at sampling sites has recorded different values ranging between 21.19 ind.100 m^{-2} (Mures, downstream Glodeni) and 502.20 ind.100 m^{-2} (Nadasa, downstream Chiheru de Jos). Other sampling sites where a high value of numerical stock was recorded, are Fitcau, downstream Alunis (494.19 ind.100 m^{-2}), Ripa, downstream Ripa de Jos (371.25 ind.100 m^{-2}), Lutul, downstream Breaza (368.02 ind.100 m^{-2}), and Beica, downstream Beica de Jos (366.64 ind.100 m^{-2}) (Fig. 1).

Weight stock

The weight stock at sampling sites has recorded the highest value in Lechinta, upstream Lechinta (3,760.06 g.100 m^{-2}), and the lowest value in Mures, downstream Glodeni (136.72 g.100 m^{-2}). Other sampling sites with an weight stock over 2,000.0 g.100 m^{-2} are Nadasa, downstream Chiheru de Jos (2,223.53 g.100 m^{-}

^{2}), Lutul, upstream Voivodeni (2,211.36 g.100 m^{-2}), Ripa, downstream Ripa de Jos (2,143.40 g.100 m^{-2}), Nirajul, upstream Eremitu (2,104.68 g.100 m^{-2}), Mures, Vidrasau side (2,058.88 g.100 m^{-2}), and Mures, upstream Brancovenesti in 2009 (2,027.92 g.100 m^{-2}) (Fig. 2).

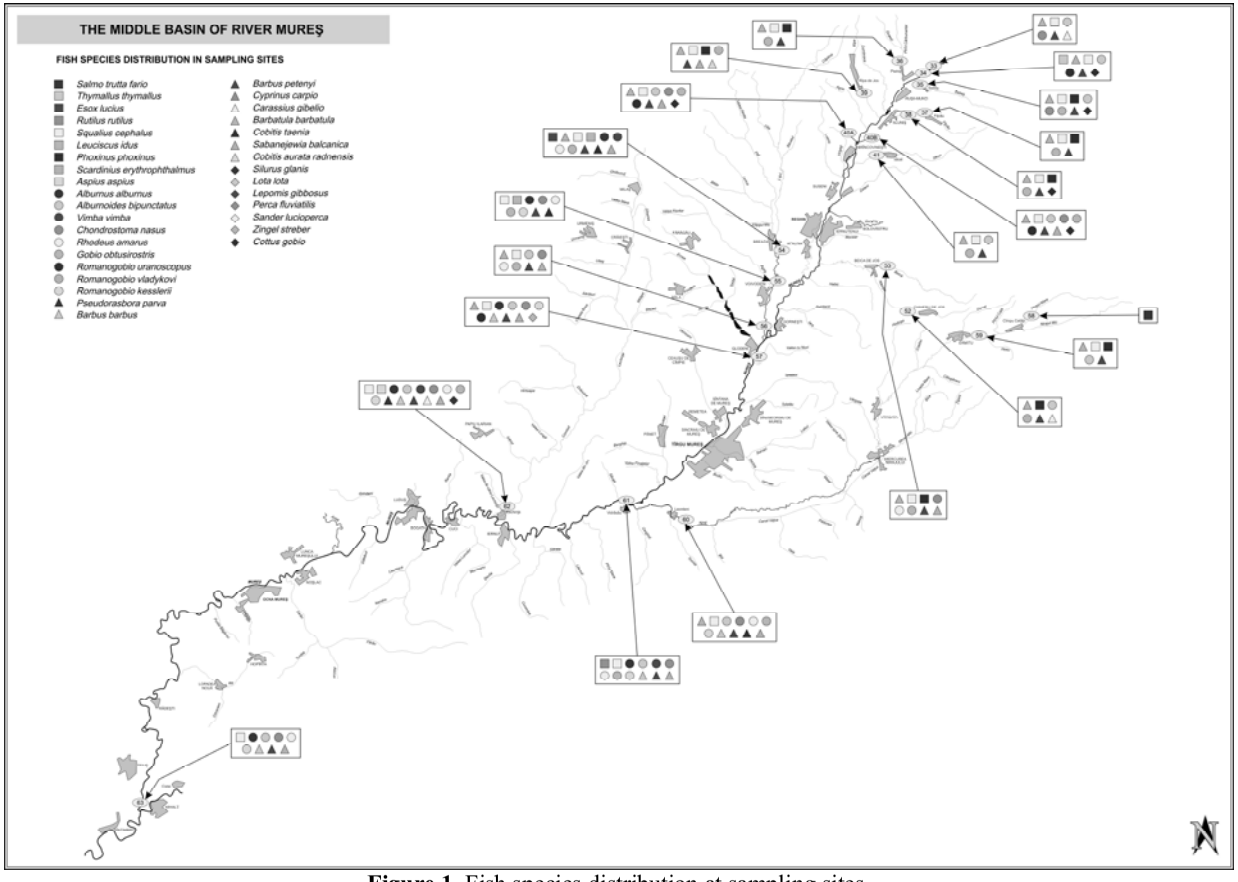


Figure 1. Fish species distribution at sampling sites.

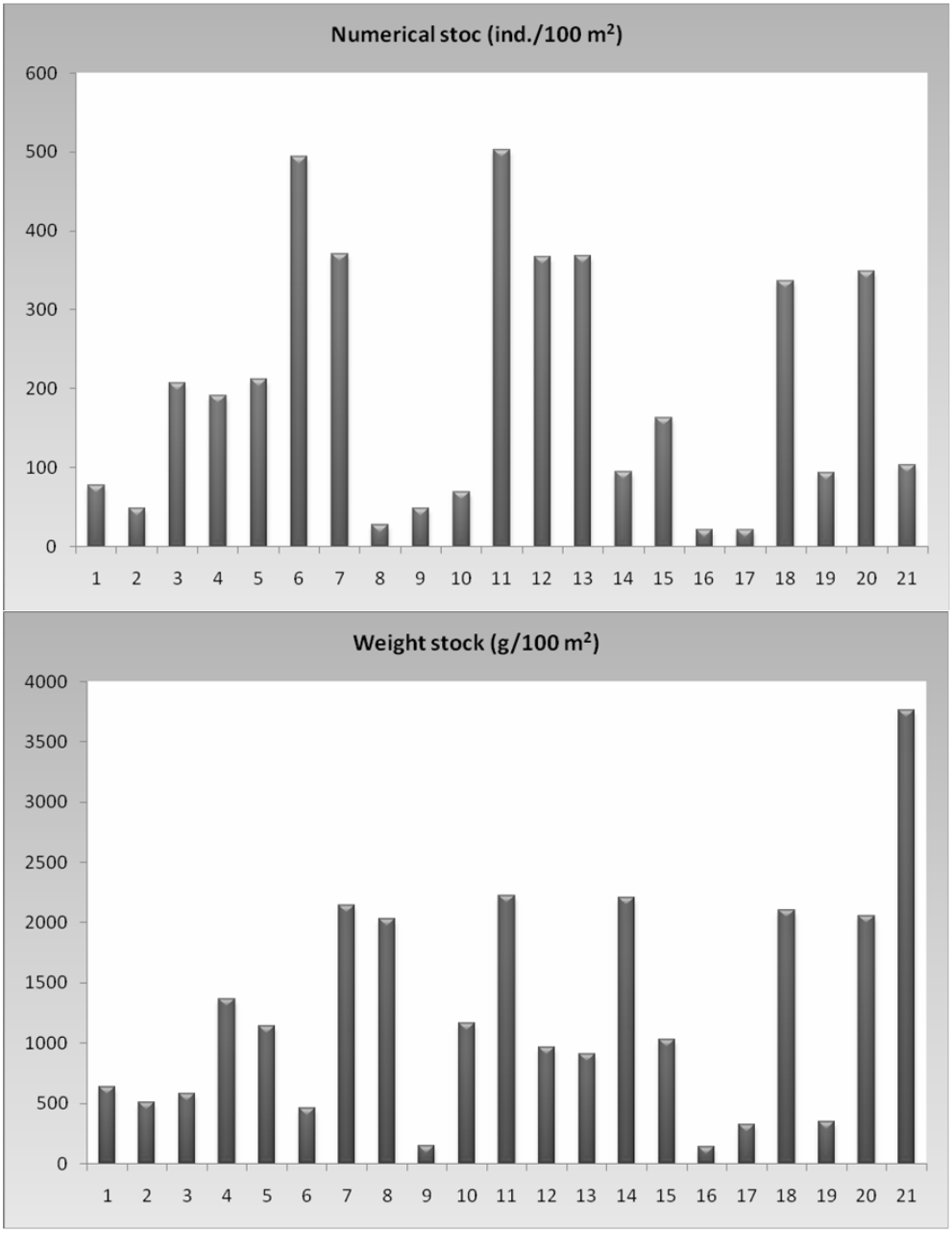


Figure 2. Numerical stock (top) and weight stock (bottom) at sampling sites in the middle basin of River Mures: 1-Mures downstream Sebes; 2-Mures confluence with Sebes; 3-Sebes confluence with Mures, downstream Sebes; 4- Paraul Carbunarilor, Pieris; 5-Fitcau downstream Fitcau; 6-Fitcau upstream Alunis; 7-Ripa downstream Ripa de Jos; 8-Mures

upstream Brancovenesti 2009; 9- Mures upstream Brancovenesti 2011; 10-Idicel downstream Idicel; 11-Nadasa downstream Chiheru de Jos; 12-Beica downstream Beica de Jos; 13-Lutul downstream Breaza; 14-Lutul upstream Voievodeni; 15-Lutul upstream confluence with Mures; 16-Mures downstream Glodeni; 17-Nirajul Mare upstream Campul Cetatii; 18-Nirajul upstream Eremitu; 19-Niraj upstream Leordeni; 20-Mures, Vidrasau side; 21-Lechinta upstream Lechinta.

Fish zones

In the middle basin of River Mures, three fish zones were revealed: brown trout zone, Mediterranean barbel zone, and chub zone. Unlike the situation in the past, the fish zones are changed. Thus, the common nase zone was replaced by the Mediterranean barbel zone in the upper part and by the chub zone in the lower part of the area.

It was noticed the lack of the leading species in the brown trout zone, as well as in the mediterranean barbel zone. It should be noted the presence of chub and stone loach as random species in brown trout zone (Fig. 3).

The biodiversity index

The biodiversity index (Shannon – Wiener) values oscillate between 0 and 2.03. The highest values of the biodiversity index were recorded in the main course of River Mures, while the lowest values were recorded in the most of its tributaries, due to the river size and consequently due to environmental conditions. In most of the sampling sites, the overall state of the fish communities is good, expressed by a biodiversity exceeding 1.00 (Fig. 4).

The biological integrity index (IBI)

The biological integrity index (IBI) at sampling sites was set by using the method of Karr & Dudley, adapted for medium and small size rivers. The assessment of fish communities' state was made according to the integrity classes.

The first four integrity classes show that fish communities are low affected, with a good self-support capacity.

The following two integrity classes indicate disturbance in structure and functionality of fish communities, while the last three integrity classes show that the fish communities are highly affected in their structure and functionality.

In this case, a monitoring program is required for habitats recovery and restoration, introduction of species, etc.

The biological integrity index (IBI) was determined for each sampling. The evaluation score was calculated based on the parameters related to species richness, trophic groups and quantitative parameters of fish communities at sampling sites.

Undoubtedly, some of the changes in the structure of fish communities are due to increasing of the human impact. However, the fish populations still have a good self-support capacity.

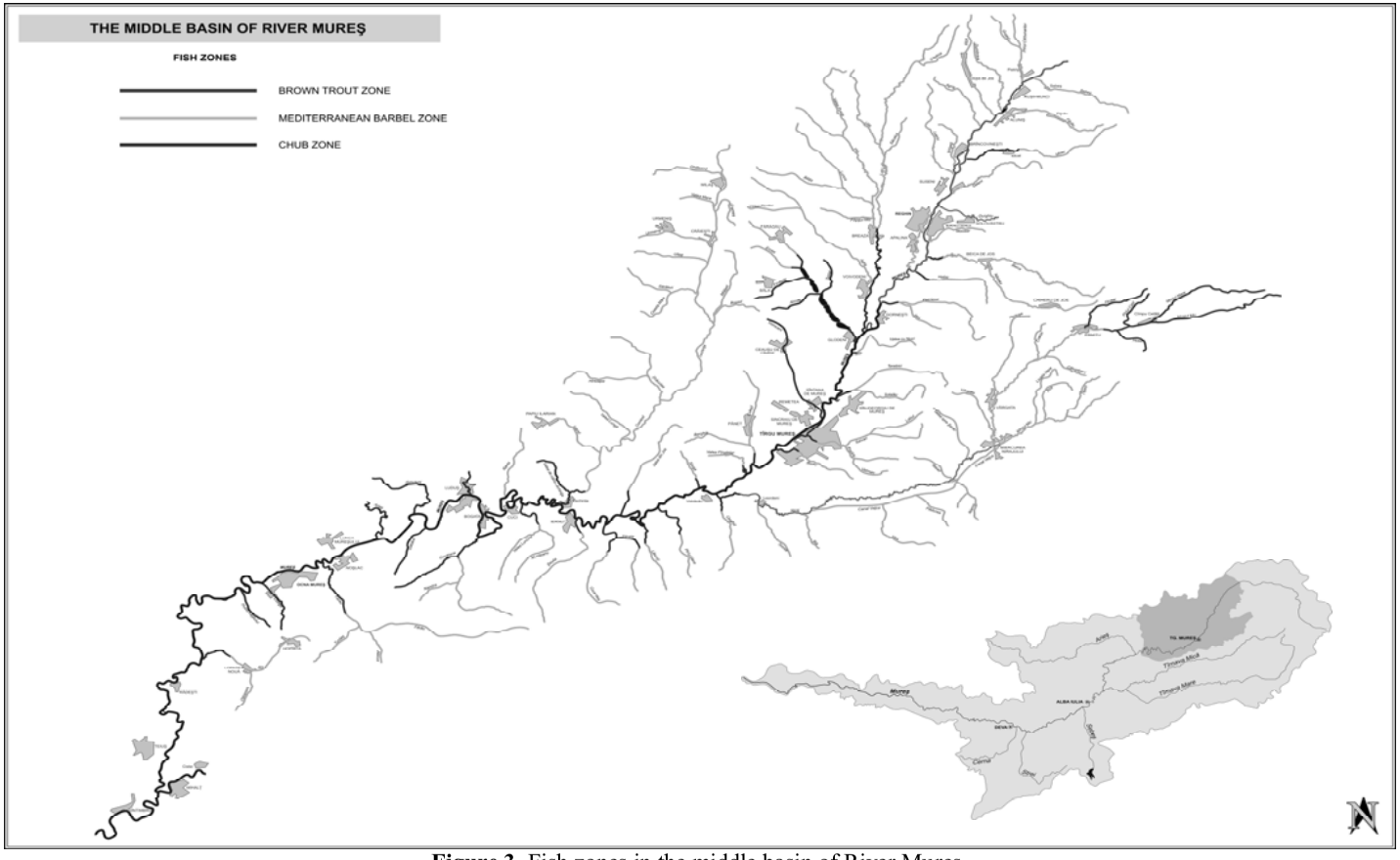


Figure 3. Fish zones in the middle basin of River Mures.

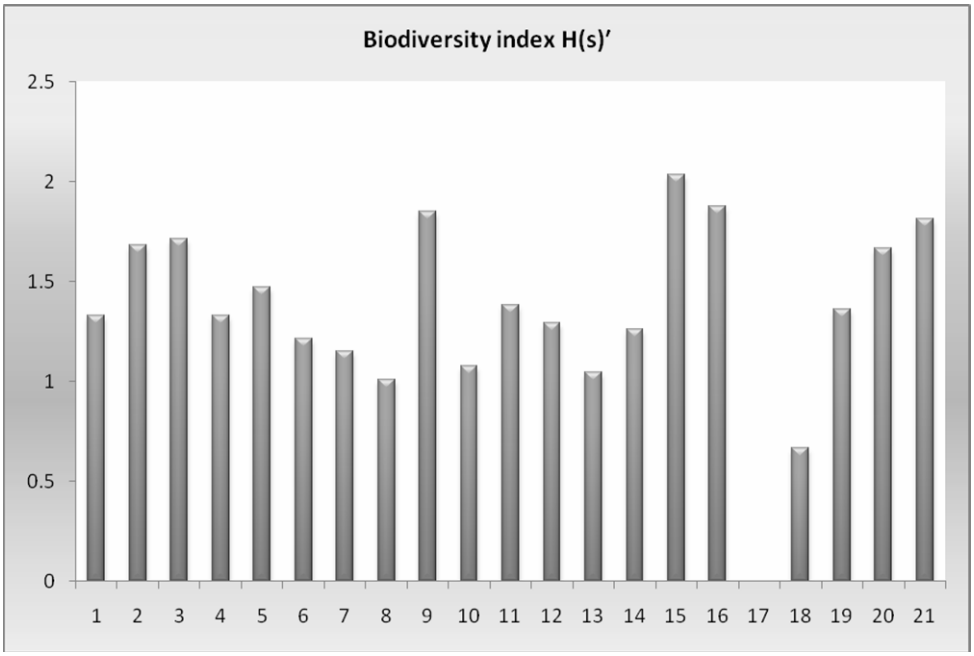


Fig. 4. Biodiversity in sampling sites in the middle basin of River Mures:
1-Mures downstream Sebes; 2-Mures confluence with Sebes; 3-Sebes confluence with Mures, downstream Sebes; 4- Paraul Carbunarilor, Pieris; 5-Fitcau downstream Fitcau; 6-Fitcau upstream Alunis; 7-Ripa downstream Ripa de Jos; 8-Mures upstream Brancovenesti 2009; 9-Mures upstream Brancovenesti 2011; 10-Idicel downstream Idicel; 11-Nadasa downstream Chiheru de Jos; 12-Beica downstream Beica de Jos; 13-Lutul downstream Breaza; 14-Lutul upstream Voievodeni; 15-Lutul upstream confluence with Mures; 16-Mures downstream Glodeni; 17-Nirajul Mare upstream Campul Cetatii; 18-Nirajul upstream Eremitu; 19-Niraj upstream Leordeni; 20-Mures, Vidrasau side; 21-Lechinta upstream Lechinta.

Conclusions

Over the period 2009-2011, in the middle basin of River Mures, 27 fish species were found, with an amount of 5,256 individuals and 32,438.6 g.

The numerical stock at sampling sites recorded values between 21.19 ind.100 m^{-2} and 502.20 ind.100 m^{-2}, while the weight stock ranged between 3,760.06 g.100 m^{-2}, and 136.72 g.100 m^{-2}.

In the middle basin of River Mures, three fish zones were revealed: brown trout zone, Mediterranean barbel zone, and chub zone. Unlike the situation in the past, the common nase zone was replaced by the Mediterranean barbel zone in the upper part and by the chub zone in the lower part of the area.

The biodiversity index (Shannon – Wiener) values oscillate between 0 and 2.03, and the highest values of the biodiversity index were recorded in the main course of River Mures.

The biological integrity index (IBI) was determined for all of the samples. The evaluation score was calculated based on the parameters related to species

richness, trophic groups and quantitative parameters of fish communities at sampling sites.

Changes in the composition of fish communities are due to the extension of the spreading area of some of the most resistant species. At the same time, some of the most sensitive fish species have special ecological requirements and a more limited area of distribution. Undoubtedly, some of the changes in the structure of fish communities are due to increasing of the human impact. However, the fish populations still have a good self-support capacity.

References

Bănăduc, D., 2005. Fish associations – habitats quality relation in the Târnave rivers ecological assesment, *Transylvanian Review of Systematical and Ecological Research*, **2**, Ed. Univ. „Lucian Blaga", Sibiu, 123-136.

Bănărescu P., 1964 – *Fauna R.P.R., Pisces-Osteichthyes, XIII*, Ed. Acad., Bucureşti.

Bănărescu, P., 2005. Ichthyological investigations in the drainage area of the Mureş River, 1948-1997, *Transylv. Rev. Syst. Ecol. Res*. 2, The Tarnava River basin, 137-144.

Froese, R., Pauly, D. (Eds.), 2011 – *Fish Base. World Wide Web electronic publication*. www.fishbase.org, version (08/2011).

Karr, J. R., 1997. *Measuring biological integrity, în Principles of conservation biology,* ed. G. K. Meffe and C. R. Carroll, Sunderland, MA: Sinauer, 483-485.

Karr, J. R., Fausch, K. D., Angermeier, P. L., Yant, P. R., Schlosser, I. J., 1986. Assessing biological integrity in running waters: a method and its rationale. Illinois Natural History Survey, Special Publication **5**.

Kottelat, M., Freyhof, J., 2007. *Handbook of European Freshwater Fishes*, Kottelat, Cornol, Switzerland and Freyhof, Berlin, Germany.

Nalbant, T., 1995. *Fish of the Mures (Maros) river: systematics and ecology*, *Tiscia monograph series*, The Maros/Mures River Valley, Szolnok, Szeged, Târgu Mureş, 225-235.

Nalbant, T., 2003. *Checklist of the fishes of Romania. Freshwater and brackishwater fishes*, *Stud. şi cerc. şt., Biologie, serie nouă, Univ. Bacău*, **8**: 122-127.

Pintilieasa, R., Ureche, D., Ureche, C., 2011. *Status of the fish communities in the upper basin of River Mures in 2009***,"** Universitatea V. Alecsandri" din Bacău, *Studii şi Cercetări Ştiinţifice, Biologie, serie nouă,* **20**/2: 158-165.

Telcean, I., 2001. Comunitatea piscicolă din cursul superior al râului Mureş, *Analele Ştiinţifice ale USMF* Nicolae Testemiţanu, Chişinău, **I**: 29-32.

Telcean, I., Cupşa, D., 2009. Fishfauna from the lowland Mureş river and the floodplain natural park area, *Analele Universitatii din Oradea, Fascicula Biologie*, **XVI**/1: 131-135.

Ujvári, I., 1972. *Geografia apelor României*, Ed. Ştiinţifică, Bucureşti.

Ureche, D., Battes, K. W., Stoica, I., 2007. Icthyofauna actual state in the upper and mid course of the River Argeş hydrographical basin, *Analele Ştiinţifice ale Univ. "Al.I. Cuza"Iaşi, secţiunea I Biologie animală*, Ed. Univ. "Al.I. Cuza"Iaşi, Tom **LIII**: 73-82.

Ureche, D., Pricope, F., Battes, K. W., 2006. A prospective monitoring of natural and anthropical fish populations in the basin of the Trotus River, Scientific Annals of the Danube Delta Institute, Tulcea, **12**: 121-130.

***1992. *Atlasul cadastrului apelor din România. Partea 1 – Date morfo-hidrografice asupra reţelei hidrografice de suprafaţă, Ministerul Mediului*, Bucureşti.

RESEARCHES ON THE FISH COMMUNITIES
IN THE BASIN OF RIVER TARNAVA MICA IN 2009

DOREL URECHE[1], RAMONA PINTILIEASA[2] and CAMELIA URECHE[1]

[1] University "Vasile Alecsandri" of Bacau, Marasesti street, no. 157, 600115, Bacau, Romania,
e-mail: dureche@ub.ro, urechec@ub.ro

[2] University "Alexandru Ioan Cuza" of Iasi, Bd. Carol I, 20A, 700505, Iasi, Romania,
e-mail: bio06ramona@yahoo.com

Abstract. The study was carried out on the main course of River Tarnava Mica and on some of its tributaries over 2009. Some of the physico-geographical and hydrochemical parameters were determined. The aim of the study was to assess the state of fish communities in the basin of River Tarnava Mica, and also to highlight significant changes in fish communities, such as biodiversity, stocks, biological integrity. The biological material was collected by electrofishing from 22 sampling sites. Over the year 2009, 20 fish species were found, with an amount of 2,035 individuals and 22,697,5 g. One of the 20 fish species is a non-native species (*Pseudorasbora parva*) and 19 are native species. We found that the numerical stock at sampling sites was between 5.14 and 361.53 ind.100 m^{-2} meanwhile the weight stock was between 90 and 1,503.38 g.100 m^{-2}. Although a high value of the numerical stock is not always correlated with a high value of the weight stock, at Domaid, upstream the confluence with Tarnava Mica we found the highest value for both of numerical and weigh stock and it is due to the large-size species (e.g. chub). Based on the ecological indices, three fish zones were found in the basin of the River Tarnava Mica: brown trout zone, mediterranean barbel zone and chub zone. Unlike the situation in the past, the fish zones are changed. Thus, the common nase zone was replaced by the mediterranean barbel zone and the barbel zone was replaced by the chub zone.

Key words: fish communities, biodiversity, biological integrity, River Tarnava Mica

Rezumat. Cercetări asupra comunităților piscicole din bazinul râului Târnava Mică în 2009. Studiul s-a desfășurat în anul 2009 pe cursul principal al râului Târnava Mică și pe unii afluenți. Au fost determinați unii parametri fizico-geografici și hidrochimici. Scopul studiului a fost de a estima starea comunităților de pești în bazinul râului Târnava Mică și de a evidenția modificările semnificative ale acestora (biodiversitate, stocuri, integritate biologică). Au fost identificate 20 specii de pești cu un total de 2035 indivizi și o biomasă de 22697,5 g. Una din cele 20 de specii este non-nativă (*Pseudorasbora parva*) iar 19 sunt native. Stocul numeric a avut valori cuprinse între 5,14 și 361,53 ind.100 m^{-2}, iar cel gravimetric între 90 și 1503,38 g.100 m^{-2}. Deși o valoare mare a stocului numeric nu este corelată întotdeauna cu o valoare mare a stocului gravimetric, pe afluentul Domaid, amonte confluența cu Târnava Mică a fost înregistrată cea mai mare valoare atât pentru stocul numeric, cât și pentru cel gravimetric și ea se datorează speciilor de talie mare, cum este cleanul. Pe baza indicilor ecologici, în bazinul râului Târnava Mică au fost identificate trei zone piscicole: zona păstrăvului, zona moioagei și zona cleanului. Spre deosebire de situația din trecut, zonele piscicole sunt modificate, astfel că zona scobarului a fost înlocuită de zona moioagei iar zona mrenei a fost înlocuită de zona cleanului.

Cuvinte cheie: comunități piscicole, biodiversitate, integritate biologică, Râul Târnava Mică

Introduction

Tarnava Mica has 191 km length and it is situated in the middle basin of the River Mures. Its springs are in Gurghiu Mountains. From its confluence with River Tarnava Mare arises River Tarnava. Near Sovata, River Tarnava Mica it recieves tributaries on the right hand only (Iuhod, Sovata) and downstream Sangeorgiu de Padure it recieves tributaries almost on the left hand only (Ujvári, 1972).

The study was carried out in the basin of River Tarnava Mica, i.e. on the main course of River Tarnava Mica and on some of its tributaries over the year 2009.

The aim of the study was to assess the current state of fish communities in this basin, and also to highlight significant changes in fish communities, such as biodiversity, stocks, biological integrity.

Material and methods

The biological material was collected by electrofishing from 22 sampling sites in 2009. The number of sampling sites was set to include all fish communities and also the main changes in the spatial distribution of the fish species. Also, the number of sampling sites was statistically ensured for the accuracy of the results. The study included qualitative and quantitative estimations. Thus, after determining the fish species, we have estimated the numerical stock (expressed in number of individuals per 100 square meters) and the weight stock (expressed in grams per 100 square meters). Some ecological indices (the index of biodiversity, the index of biological integrity) and some of the physico-geographical and hydrochemical parameters were also determined.

Results and discutions

Our study, based on the ecological analysis, revealed some important differences compared with the situation in the past (Banarescu, 1964).

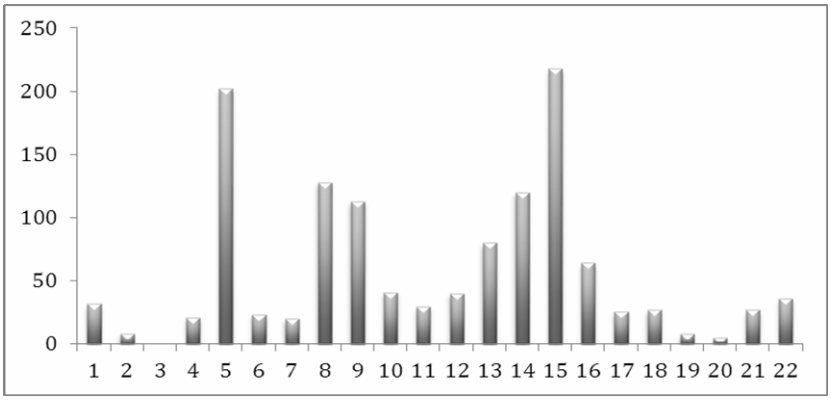


Figure 1. Numerical stock at sampling sites (ind.100 m^{-2}):
1-Creanga Mare upstream Praid; 2-Tarnava Mica upstream Praid; 3-Corund 4.5 km upstream Corund; 4- Corund upstream 2 km Corund; 5-Corund upstream Ocna de Sus; 6-Iuhod downstream Ilieşi; 7-Sovata downstream Capeti; 8-Tarnava Mica downstream Sovata; 9-Tarnava Mica upstream confluence with Solocma; 10-Tarnava Mica downstream bridge Sangeorgiu de Padure; 11-Gheahes downstream Neaua; 12-Tarnava Mica upstream Fantanele; 13-Vetca downstream Bordosiu; 14-Nades upstream Chedru; 15-Domaid upstream confluence with Tarnava Mica; 16-Tarnava Mica upstream Soimus; 17-Tarnava Mica downstream Suplac; 18-Tarnava Mica downstream Mica; 19-Tarnava Mica upstream Adamus; 20-Tarnava Mica downstream Jodvei; 21-Tarnava Mica upstream Panade bridge; 22-Tarnava Mica downstream Blaj.

Over the year 2009, 20 fish species were found, with an amount of 2,035 individuals and 22,697.5 g.

The numerical stock at sampling sites ranged between 217.59 ind.100 m^{-2} (Domaid, upstream confluence with Tarnava Mica) and 5.14 ind.100 m^{-2} (Tarnava Mica, downstream Jidvei) (Fig. 1). A high value of the numerical stock was also found in Corund, upstream Ocna de Sus (201.75 ind.100 m^{-2}).

The weight stock at sampling sites ranged between 1,066.97 g.100 m^{-2} (Tarnava Mica, upstream Panade bridge), and 90 g.100 m^{-2} (Gheahes, downstream Neaua) (Fig. 2).

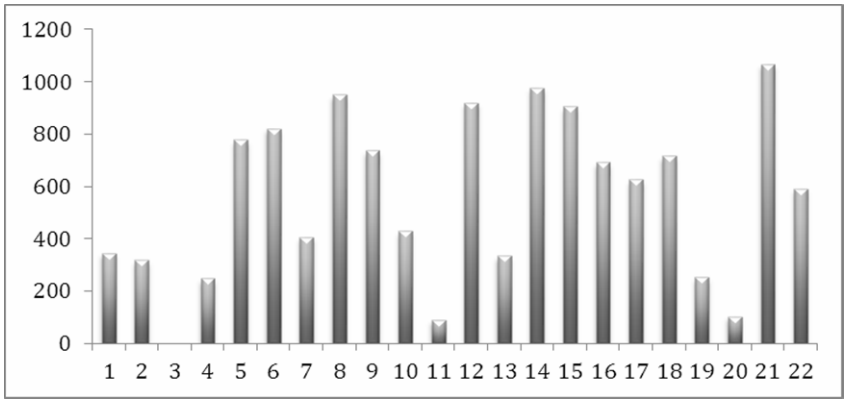


Figure 2. Weight stock at sampling sites (ind.100 m^{-2}):
1-Creanga Mare upstream Praid; 2-Tarnava Mica upstream Praid; 3-Corund 4.5 km upstream Corund; 4- Corund upstream 2 km Corund; 5-Corund upstream Ocna de Sus; 6-Iuhod downstream Ilieși; 7-Sovata downstream Capeti; 8-Tarnava Mica downstream Sovata; 9-Tarnava Mica upstream confluence with Solocma; 10-Tarnava Mica downstream bridge Sangeorgiu de Padure; 11-Gheahes downstream Neaua; 12-Tarnava Mica upstream Fantanele; 13-Vetca downstream Bordosiu; 14-Nades upstream Chedru; 15-Domaid upstream confluence with Tarnava Mica; 16-Tarnava Mica upstream Soimus; 17-Tarnava Mica downstream Suplac; 18-Tarnava Mica downstream Mica; 19-Tarnava Mica upstream Adamus; 20-Tarnava Mica downstream Jodvei; 21-Tarnava Mica upstream Panade bridge; 22-Tarnava Mica downstream Blaj.

Comparing the numerical stock at sampling sites, we found that it is significantly increased at two of the sampling sites (Domaid, upstream confluence with Tarnava Mica; Corund, upstream Ocna de Sus), due to a high number of individuals of chub (*Squalius cephalus*) and schneider (*Alburnoides bipunctatus*). The numerical stock at this sampling site is correlated with an increased weigh stock due to the large-sized species (chub).

However, a decrease in numerical stock is not always correlated with a decrease in weight stock. This is the situation found in some of the sampling sites, where a decreased numerical stock is correlated with an increased weight stock due to the large-sized species (e.g. chub, in most of cases).

It was found that the biodiversity has remained unchanged in 2009 (20 species), compared to the situation occurred in the last decades of the 20th century (20 species) (Banarescu, 1964; Nalbant, 2003).

Although, it was noticed the lack of a number of 4 species of the 20 found by Banarescu (1964) in the area where the study was carried out in 2009 (*Romanogobio vladykovi, Misgurnus fossilis, Cyprinus carpio* and *Thymallus thymallus*).

On the other hand, over the year 2009, five new fish species were found in addition to situation recorded by Banarescu in 1964, respectively *Aspius aspius, Vimba vimba, Pseudorasbora parva, Cobitis taenia* and *Silurus glanis.*

Biodiversity at the sampling sites is variable. We found that the highest biodiversity in 2009 was on the River Tarnava Mica, downstream Sangeorgiu de Padure bridge, downstream Suplac and downstream Blaj, and on the River Domaid, upstream confluence with Tarnava Mica (9 species).

Concerning the fish zones, in the last decades of the 20th century, Banarescu has described 4 fish zones, from springs to river mouth: brown trout zone, grayling zone, common nase zone and barbell zone.

In our study, based on the ecological indices, three fish zones were found in the basin of the River Tarnava Mica: brown trout zone, mediterranean barbel zone and chub zone (Fig. 6). As we can see, the grayling zone has been replaced by the mediterranean barbel zone, and the last two fish zones (common nase zone and barbell zone) have been replaced by the chub zone.

The numerical stock and the weight stock at the sampling sites of the three fish zones are represented in Fig. 3-5.

We can observe that at some of the sampling sites, a high value of numerical stock is correlated with a low value of the weight stock and vice-versa, due to the small-sized species in the first case and due to the large-sized species in the second case.

The index of biological integrity (IBI) (Karr, 1997) was calculated based on a set of parameters related to fish communities, and it has recorded different values at different sampling sites. At most of the sampling sites, in the upper part of the basin, the values of the index of biological integrity reveal a good integrity class. In the lower part of the basin, the index of biological integrity has recorded low values indicating a moderate integrity class. The situation found in the lower part of the basin of River Tarnava Mica is the result of the interactions of different factors, especially chemical pollution and domestic pollution. A very low integrity class was found at a single sampling site (Corund, 4.5 km upstream Corund locality) which is situated very close to springs and with shallow water (Fig. 7).

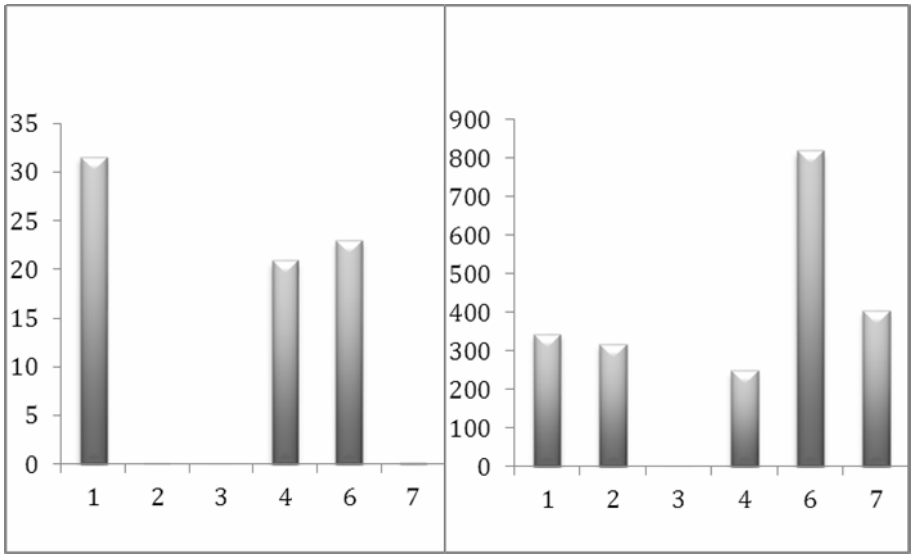


Figure 3. Numerical stock (ind.100 m^{-2}) (left) and weight stock (g.100 m^{-2}) (right) at sampling sites in brown trout zone: 1-Creanga Mare, upstream Praid; 2-Tarnava Mica, upstream Praid; 3-Corund, 4.5 km upstream Corund; 4-Corund, upstream 2 km Corund; 6-Iuhod, downstream Iliesi; 7-Sovata, downstream Capeti.

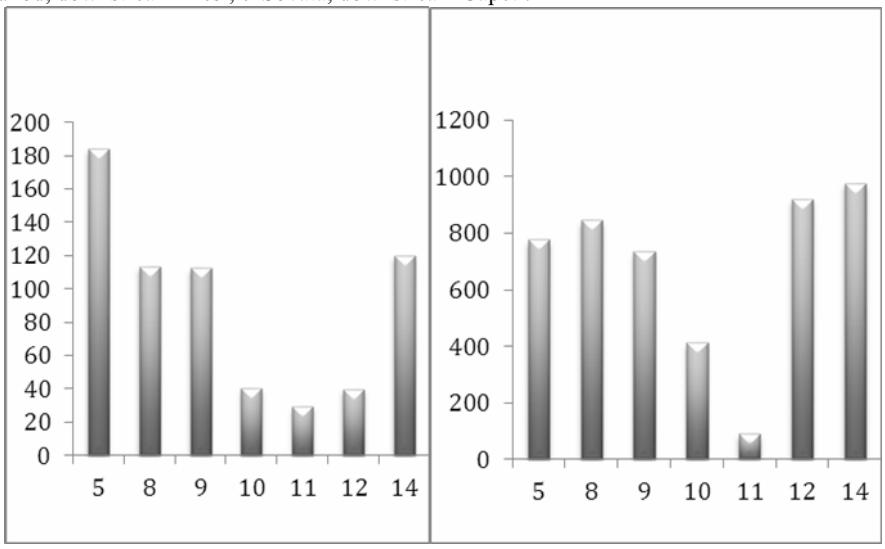


Figure 4. Numerical stock (ind.100 m^{-2}) (left) and weight stock (g.100 m^{-2}) (right) at sampling sites in mediterranean barbel zone: 5-Corund, upstream Ocna de Sus; 8-Tarnava Mica, downstream Sovata; 9-Tarnava Mica, upstream confluence with Solocma; 10-Tarnava Mica, downstream bridge Sangeorgiu de Padure; 11-Gheahes, downstream Neaua; 12-Tarnava Mica, upstream Fantanele; 14-Nades, upstream Chedru.

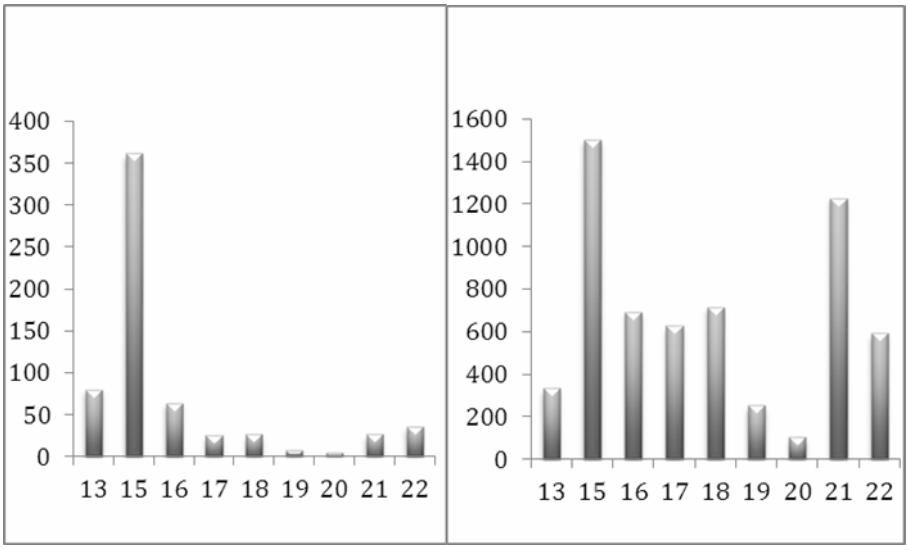


Figure 5. Numerical stock (ind.100 m^{-2}) (left) and weight stock (g.100 m^{-2}) (right) at sampling sites in chub zone: 13-Vetca, downstream Bordosiu; 15-Domaid, upstream confluence with Tarnava Mica; 16-Tarnava Mica, upstream Soimus; 17-Tarnava Mica, downstream Suplac; 18-Tarnava Mica, downstream Mica; 19-Tarnava Mica, upstream Adamus; 20-Tarnava Mica, downstream Jodvei; 21-Tarnava Mica, upstream Panade bridge; 22-Tarnava Mica, downstream Blaj.

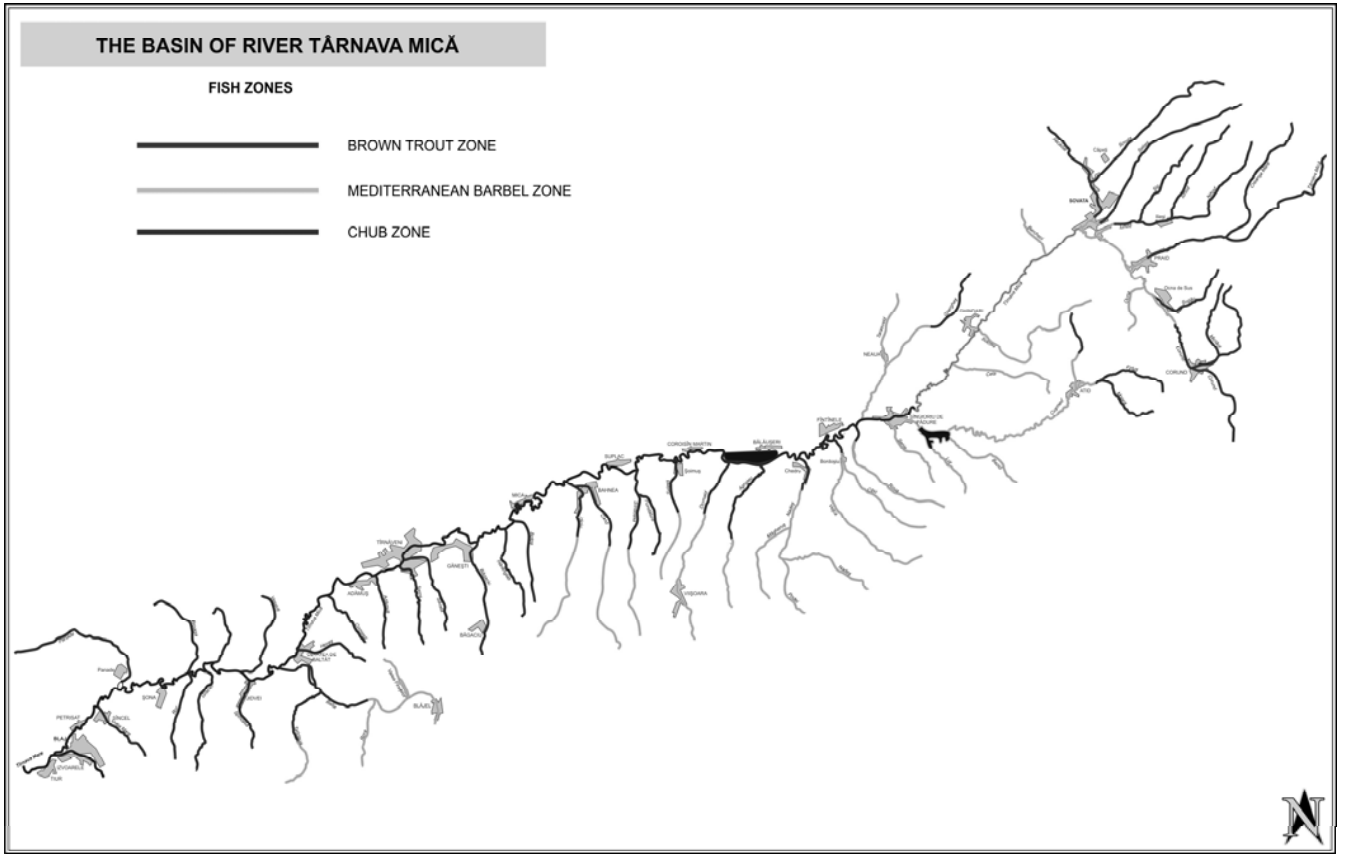


Figure 6. Fish zones in the basin of River Tarnava Mica.

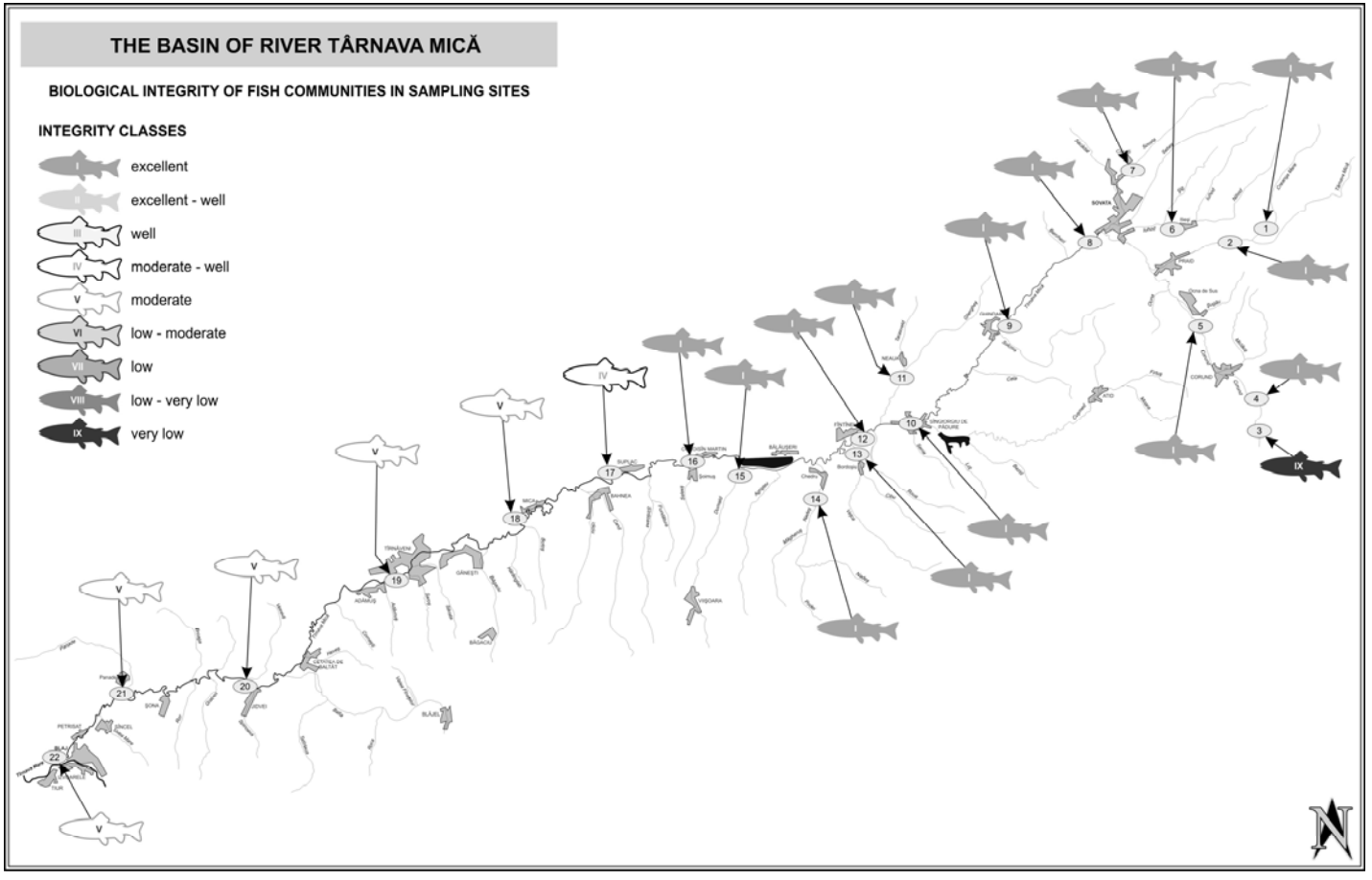

THE BASIN OF RIVER TÂRNAVA MICĂ
BIOLOGICAL INTEGRITY OF FISH COMMUNITIES IN SAMPLING SITES
INTEGRITY CLASSES
I excellent
II excellent - well
III well
IV moderate - well
V moderate
VI low - moderate
VII low
VIII low - very low
IX very low
N

Figure 7. Biological integrity of fish communities at sampling sites in the basin of River Tarnava Mica.

Conclusions

The study was carried out in the basin of River Tarnava Mica over the year 2009. Our research identified 20 species (2,035 individuals), collected by electrofishing from 22 sampling sites located on the main course of the River Tarnava Mica and some of its tributaries.

Numerical stock ranged between 217.59 ind.100 m^{-2} and 5.14 ind.100 m^{-2} while the weight stock ranged between 1066.97 g.100 m^{-2} and 90 g.100 m^{-2}.

Biodiversity index showed the presence of certain relatively well-balanced fish communities, with numerous native species.

Undoubtedly, some of the changes in fish communities structure are due to wastewater discharge from households placed in riparian areas. Other changes in the composition of fish communities are due to the extension of the spreading area of some of the most resistant species. On the other hand, some of the most sensitive fish species have special ecological requirements and a more limited area of distribution.

However, the fish populations in the basin of the River Tarnava Mica have a good self-support capacity.

References

Banarescu, P., 1964. *Fauna R.P.R., Pisces-Osteichthyes, XIII*, Ed. Acad., Bucureşti.

Froese, R., Pauly, D. (Eds.), 2011. *FishBase. World Wide Web electronic publication*. www.fishbase.org, version (08/2011).

Karr, J. R., 1997. *Measuring biological integrity, în Principles of conservation biology,* ed. G. K. Meffe and C. R. Carroll, Sunderland, MA: Sinauer, 483-485.

Kottelat, M., Freyhof, J., 2007. *Handbook of European Freshwater Fishes*, Kottelat, Cornol, Switzerland and Freyhof, Berlin, Germany.

Nalbant, T., 1995. Fish of the Mures (Maros) river: systematics and ecology, *Tiscia monograph series*, The Maros/Mures River Valley, Szolnok, Szeged, Târgu Mureş.

Nalbant, T., 2003. Checklist of the fishes of Romania. Freshwater and brackishwater fishes, *Stud. şi cerc. şt., Biologie, serie nouă*, Univ. Bacău, **8**: 122-127.

Pintilieasa, R., Ureche, D., Ureche, C., 2011. Status of the fish communities in the upper basin of River Mures in 2009**,"** Universitatea V. Alecsandri" din Bacău, *Studii şi Cercetări Ştiinţifice, Biologie, serie nouă*, **20**/2: 158-165.

Ujvári, I., 1972. *Geografia apelor României*, Ed. Ştiinţifică, Bucureşti.

Ureche, D., Battes, K. W., Stoica, I. 2007. Icthyofauna actual state in the upper and mid course of the River Argeş hydrographical basin, *Analele Ştiinţifice ale Univ. "Al.I. Cuza" Iaşi, secţiunea I Biologie animală*, Ed. Univ. "Al.I. Cuza" Iaşi, **LIII**: 73-82.

Ureche, D., Pricope, F., Battes, K. W. 2006. A prospective monitoring of natural and anthropical fish populations in the basin of the Trotus River, *Scientific Annals of the Danube Delta Institute*, Tulcea, **12**: 121-130.

THE INFLUENCE OF ENVIRONMENTAL FACTORS (TEMPERATURE, RAINFALL, SOIL MOISTURE, PH) ON THE DENSITY AND BIOMASS OF *APORRECTODEA ROSEA ROSEA* SPECIES

GHEORGHITA BRINZEA, ALINA PAUNESCU and MARIA CRISTINA PONEPAL

University of Pitesti, Faculty of Science, Targu din Vale Street, no.1, Pitesti, Romania,
e-mail: georgeta_branzea@yahoo.com; alina_paunescu@yahoo.com

Abstract. The present study regards the influence of environmental factors on the density and biomass of *Aporrectodea rosea rosea* species, which is dominant in the community of earthworms found in some forest ecosystems: deciduous forest (Ruginoasa Station), spruce forest (Cetate-Molid Station) and grassland (Cetate-Pajiste Station), belonging to the high hills of the Piedmont Cândeşti, Arges County, in the period March-October 2007. The simple correlations between the density of *Aporrectodea rosea rosea* species and the environmental factors (temperature, rain-fall, soil moisture and pH) have shown that the high temperatures in 2007 affected the numerical density of the species leading to its decrease. The rain-fall in that year had a lower impact on species density, while the influence of soil moisture and pH was insignificant. The monthly biomass of soil layers was higher in spring and autumn than in summer.

Key words: lumbricidae, *Aporrectodea*, correlations, biomass

Rezumat. Influenţa factorilor de mediu (temperatură, precipitaţii, umiditatea solului, pH) asupra densităţii şi biomasei speciei *Aporrectodea rosea rosea.* În această lucrare am realizat un studiu privind influenţa factorilor de mediu asupra densităţii şi biomasei speciei *Aporrectodea rosea rosea*, specie considerată dominantă în cadrul comunităţii de râme identificate în unele ecosisteme forestiere, respectiv o pădure de foioase (Staţia Ruginoasa), o pădure de molid (Staţia Cetate-Molid) şi o pajişte (Staţia Cetate - Pajişte) aparţinând dealurilor înalte ale Piemontului Cândeşti din Judeţul Argeş, în perioada martie-octombrie 2007. Corelaţiile simple dintre densitatea speciei *Aporrectodea rosearosea* şi factorii de mediu (temperatură, precipitaţii, umiditatea solului, pH), au demonstrat că temperaturile crescute din anul 2007 au afectat densitatea numerică a speciei determinând o scădere a acesteia. Precipitaţiile din anul respectiv au avut un impact mai redus asupra densităţii speciei, influenţa umidităţii solului şi a pH-ului a fost nesemnificativă. Biomasa lunară în straturile de sol a fost mai mare în lunile de primăvară şi toamnă comparativ cu lunile de vară.

Cuvinte cheie: lumbricide, *Aporrectodea*, corelaţii, densitate, biomasă

Introduction

Lumbricidae is a group of soil invertebrates with the most important role in the development processes of decomposition and mineralization of organic matter. Earthworm biomass, abundance and community composition is affected by various environmental and management factors, i.e. abiotic factors, such as climate, soil, vegetation and litter supply, and biotic factors, such as competition, predation, parasitism, disease and food relationship (Curry, 2004). How these parameters vary is the dynamic response of lumbricidae species to climate changes. Ecologists recognized the central role of climate, e.g., precipitation and temperature, in determining the primary production of terrestrial ecosystems (Holdridge, 1947; Lieth&Whittaker, 1975; Whittaker, 1975). Climate, predominantly temperature and

moisture conditions, is regarded to be the most important factor controlling earthworm biomass and abundance in the soil. The temperature tolerance of earthworms is narrow, generally ranging from 0-30°C with the optimum for temperate species typically being in the range of 10-20°C.

However, some tropical and subtropical species are adapted to temperatures above 30°C and there are some species reported to tolerate temperatures below 0°C. Soil temperature also determines soil moisture conditions. High temperature is often correlated with moisture shortage and moisture stress for soil animals. Earthworms depend on the chemical and physical properties of the soil, such as pH, texture, and organic matter supply. The concentration of hydrogen ions (pH) is supposed to be an important environmental factor limiting earthworm abundance; earthworm species vary in their pH tolerance and preference. *In situ* observation of the dynamics of earthworm population's biomass is a means to determine the long-term effects on the environment (Kooch & Jalivand, 2008). The burrow systems of earthworms are affected by site-specific factors such as soil texture and topography, as well as the types of earthworms present (Capowiez *et al.*, 1998; Bastardie et al., 2005), but earthworm populations respond in predictable ways to fluctuations in soil temperature and moisture. Kretzschmar (1982) reported that the seasonal variation in burrow numbers and forms in grassland was related to fluctuations in water content and temperature. A considerable amount of attention has been paid to understanding the effect of climatic variation, including drought, on aboveground production in a variety of terrestrial ecosystems (Sala *et al.*, 1988; Knapp & Smith, 2001; Knapp *et al.*, 2002). Earthworms were considered as useful in assessing heavy metal pollution in soils, because they proved to be sensitive to pollution (Malley *et al.*, 2006).

The purpose of this study was to observe the monthly dynamics of *Aporrectodea rosea rosea,* in terms of numerical density and biomass density, depending on environmental conditions (temperature, rainfall, soil moisture, and pH). This dominant species has a key role in the decomposition and mineralization processes of organic substances in the three forest ecosystems analyzed (deciduous forest, spruce forest and secondary natural grassland).

Materials and methods

The faunistic material was monthly collected, from March to October 2007. The determination of species was made in the laboratory by means of determinators (Pop, 1949; Easton, 1983). Monthly dynamics of *Aporrectodea rosea rosea* species was highlighted by the evolution of numerical density, expressed by the average number of of individuals m^{-2}. Simple correlations were made to express the influence of abiotic factors (temperature, rainfall, soil moisture, pH), on the density and biomass of lumbricidae in different soil layers from 0-10 cm (S_1) to 30-40 cm (S_3 - S4). It was calculated the simple correlation coefficient „r" and its significance, using SPSS 10,0 For Windows. The trend line equation was also calculated. The biomass was calculated by the ratio of individual and dry weight of each individual of the species (mg.d.s. m^{-2}). Individual live weight and individual weight after drying were determined by weighing on analytical balance. To remove water from the body, the faunistic material was dried in a drying cabinet, at a temperature of

105 ^{0}C. Drying was complete, when, by successive weighings, after 48 hours, the weight of the individuals remained constant.

Results and discussions

The numerical density of lumbricidae populations is a structural parameter influenced by environmental factors, sensitive to their unique mode fluctuations. The way in which this parameter varies is the dynamic response of populations to climate changes. The degree to which these factors influence the numerical density of lumbricidae is highlighted by the simple correlation analysis between numerical density of the soil in different layers (dependent variable) and one of the environmental factors (temperature, rainfall, soil moisture, pH) as an independent variable taken separately.

Data on correlations between density of *Aporrectodea rosea rosea* species and environmental factors are shown in Figs. 1-12.

The analysis of the simple correlation coefficient „r" in 2007 for *Aporrectodea rosea rosea* species in the soil layers 0-10 cm, 10-20 cm, 20-30 cm, 30-40 cm showed that high temeperatures of the year affected the numerical density of the species, leading to its decline in the three habitats studied on the whole, with a significant correlation between density and temperature, for a significance test $p<0.01$ (Fig. 1).

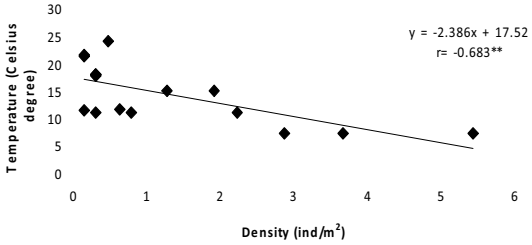


Figure 1. Correlation between density (ind.m^{-2}) and temperature (C^0) for *Aporrectodea rosea rosea* species in 2007. (** - the correlations is significant for $p<0.01$).

The rainfall had a smaller impact on the species density, as demonstrated by the correlation coefficient which was insignificant for $p<0.01$ (Fig. 2).

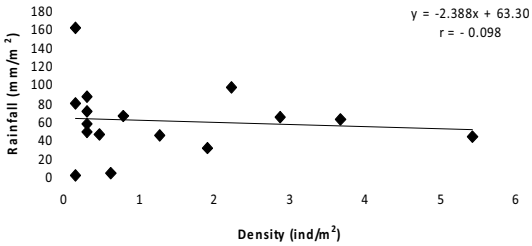


Figure 2. Correlation between density (ind.m^{-2}) and rainfall for *Aporrectodea rosea rosea* species in 2007.

The influence of soil moisture and pH on the soil layers (L, S_1 S_2 S_3 S_4) was insignificant. However, density sharply decreased in the layer 30-40 cm, with increasing pH; in this case, the correlation was significant for a significance test $p<0.05$ (Fig. 12).

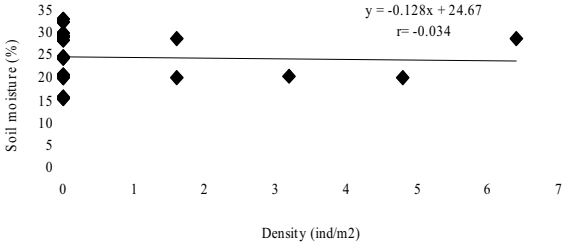


Figure 3. Correlation between density (ind.m^{-2}) and soil moisture in the litter for *Aporrectodea rosea rosea* species in 2007.

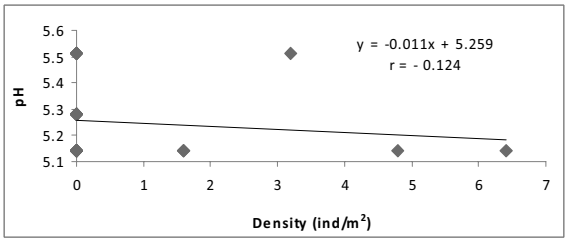


Figure 4. Correlation between density (ind.m^{-2}) and soil pH in the litter for *Aporrectodea rosea rosea* species in 2007.

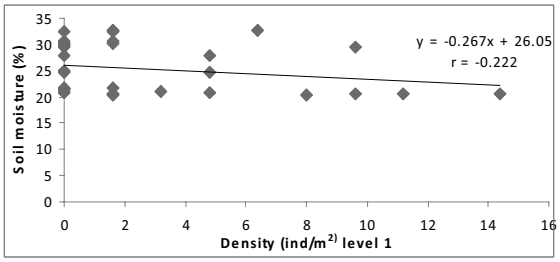


Figure 5. Correlation between density (ind.m^{-2}) and soil moisture in the layer 1 (0-10 cm) for *Aporrectodea rosea rosea* species in 2007.

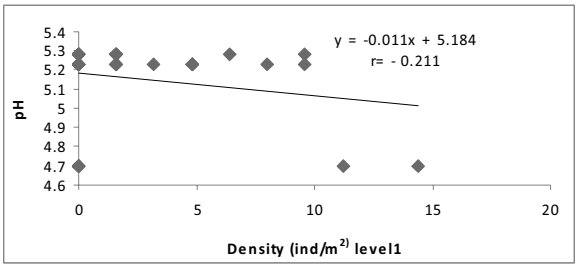


Figure 6. Correlation between density (ind.m^{-2}) and soil pH in the layer 1 (0-10 cm) for *Aporrectodea rosea rosea* species in 2007.

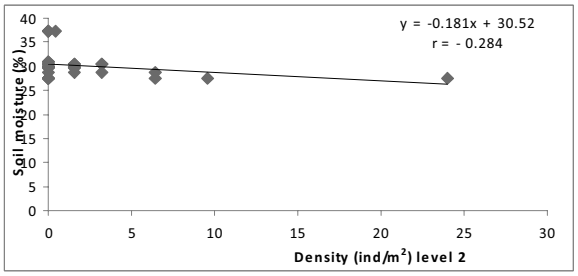


Figure 7. Correlation between density (ind.m^{-2}) and soil moisture in the layer 2 (0-20 cm) for *Aporrectodea rosea rosea* species in 2007.

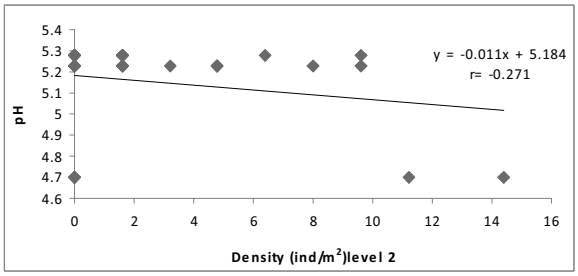


Figure 8. Correlation between density (ind.m^{-2}) and soil pH in the layer 2 (0-20 cm) for *Aporrectodea rosea rosea* species in 2007 (** - the correlations is significant for $p<0.01$).

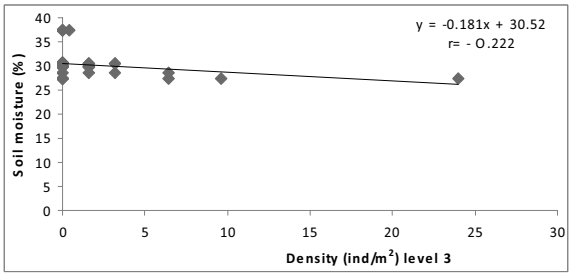


Figure 9. Correlation between density (ind.m^{-2}) and soil moisture in the layer 3 (20-30 cm) for *Aporrectodea rosea rosea* species in 2007.

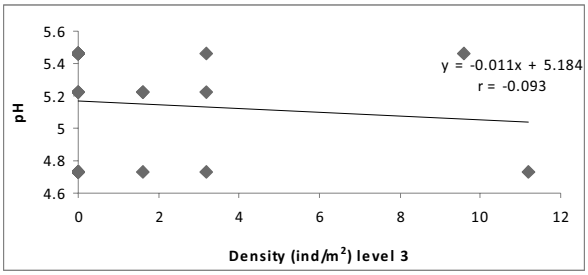


Figure 10. Correlation between density (ind.m^{-2}) and soil pH in the layer 3 (20-30 cm) for *Aporrectodea rosea rosea* species in 2007 (** - the correlations is significant for $p<0.01$) .

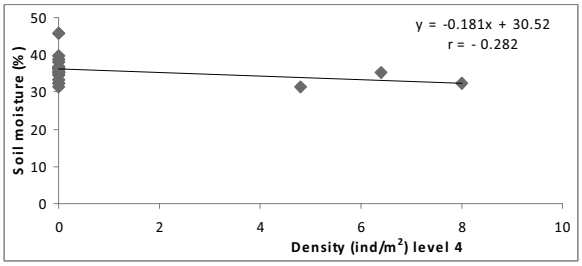


Figure 11. Correlation between density (ind.m^{-2}) and soil moisture in the layer 4 (30-40 cm) for *Aporrectodea rosea rosea* species in 2007.

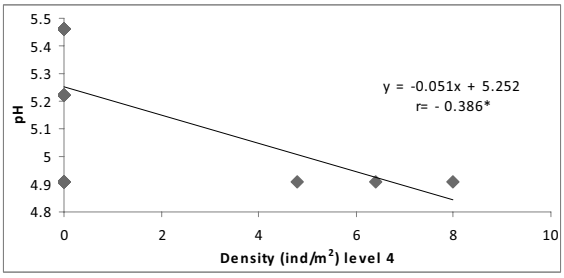


Figure 12. Correlation between density (ind.m^{-2}) and soil pH in the layer 4 (30-40 cm) for *Aporrectodea rosea rosea* species in 2007 (** - the correlations is significant for $p<0.05$).

The soil properties (pH-values, humidity, Corg-content, soil texture) and the type of land use are suggested as the determining factors for the occurrence of different earthworm species. The abundance and biomass are suitable indicators for the classification of land use for specific earthworm communities. The developed matrix for expected occurrence of earthworm species can help to achieve an easy assessment of different habitats.

The structure of earthworm community is sensitive to particular physico-chemical soil properties (Decaens *et al.*, 2003). Many authors refer to the influence of pH-value (Nordstrom & Rundgren, 1974; Satchell, 1983; Curry, 1998; Paoletti, 2001; Graefe & Beylich, 2003), humus content and soil humidity on the occurrence

of earthworms (Nordstrom & Rundgren, 1974; Satchell, 1983; Lee, 1985; Rombke *et al.*, 2000; Sommer *et al.*, 2002; Whalen & Costa, 2003). Soil moisture influences the abundance of earthworm communities more than soil type (Ivask *et al.*, 2006). Of course, the dependency relations between lumbricidae and abiotic environmental factors are not always easily expressed in mathematical relations. Understanding the dependency relations between abiotic factors and lumbricidae only by simple correlations, does not surprise the relation complexity, the influence that these closely interdependent factors exercises on lumbricidae populations.

Fig. 13 shows the monthly dynamics of the biomass for *Aporrectodea rosea rosea* species, during March-October 2007, in the five soil layers of the ecosystems analysed. The species, considered eudominant in terms of numerical density, in the deciduous forest ecosystem (Ruginoasa Station), recorded a biomass of 0.641 mg.d.s. m^{-2} in March, in the layer 0-10 cm (S1), with decreasing values in the layer 20-30 cm (S3) (0.187 mg.d.s. m^{-2}). In May, it recorded very low values (0.099 mg.d.s. m^{-2}) in the litter. The same happened in October in the layers S1, S2, S3 (from 0.009 to 0.012 mg.d.s. m^{-2}). The biomass was very low in this ecosystem. Increases of the biomass were observed in spring and autumn months in the layers S1, S2, S3, with values between 0.01 – 0.413 mg.d.s. m^{-2}, in the spruce forest (Cetate-Spruce Station).

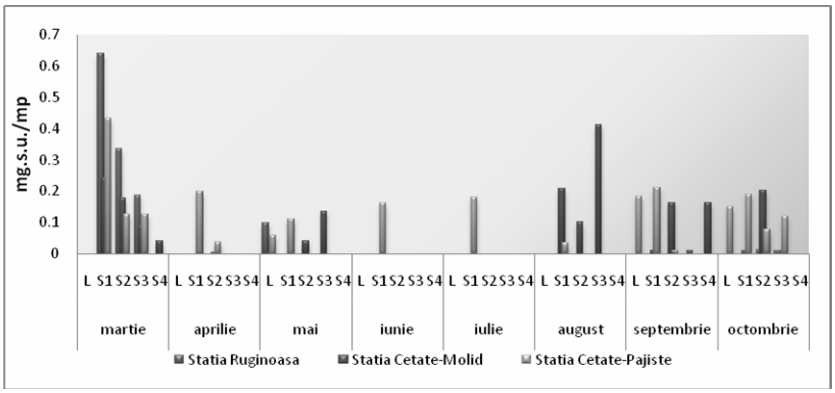


Figure 13. Monthly dynamics of the biomass for *Aporrectodea rosea rosea* species in the soil layers during March-October 2007.

The grassland (Cetate – Grassland Station) recorded increases of the biomass especially in the layer S1 (10 cm), from 0.034 to 0.432 mg.d.s. m^{-2}. In autumn months, the amount of litter in the following layers was reduced. In March, the highest biomass was recorded in the layers 0-10 cm (S_1= 0.432 mg.d.s. m^{-2}), 10-20 cm (S_2= 0.124 mg.d.s. m^{-2}) and 20-30 cm (S_3= 0.125 mg.d.s. m^{-2}). The biomass values in this ecosystem ranged from 0.11 to 0.432 mg.d.s. m^{-2}. The analysis of the biomass for *Aporectodea rosea rosea* species in the five soil layers of the ecosystems analysed in 2007 showed values lower than 1mg.d.s. m^{-2}.

Total biomass of *Aporrectodea rosea rosea* species is shown in Fig. 14. It was found out that the highest value of total biomass was recorded in March (1.163

mg.d.s. m^{-2}), representing 90% of the biomass in the deciduous forest ecosystem (Ruginoasa Station).

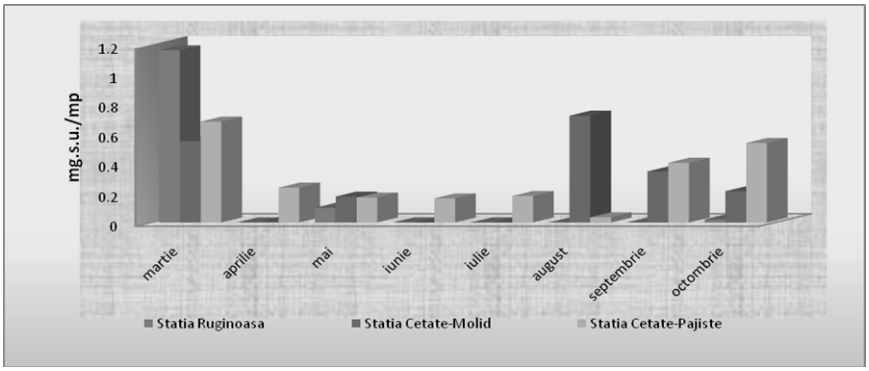


Figure 14. Monthly total biomass of *Aporrectodea rosea rosea* species during March-October 2007.

In the spruce forest (Cetate-Spruce Station), the highest value of the biomass was recorded in August (0.721 mg.d.s. m^{-2}), representing 35.97% of total monthly biomass. Increases of the biomass in March were also observed in the grassland, with a value of 0.681 mg.d.s. m^{-2}, representing 28.36% of total biomass. Total biomass value in the grassland was higher in March, April, September, October and relatively constant for the rest of the period.

Conclusions

Temperature increase in 2007 resulted in a decrease of numerical density for *Aporrectodea rosea rosea* species; the low rainfall also had an important role in the dynamics of numerical density; pH increase over 5.3 caused a decrease in density, at a significance threshold $p<0,05$; excessive soil moisture in the grassland, due to a nearby watercourse, also caused a decrease in density of this species, the ecosystem soil water decreasing the amount of oxygen in the soil. The biomass obtained in the three ecosystems, depended chiefly on the monthly density of the species and equally on the individual biomass, knowing that the species was smaller. Also, increasing the number of individuals within the species, has increased the total biomass. Since metabolism and activity of lumbricidae decreased in summer months, due to less moisture in the soil layers, caused a decrease in total and individual biomass.

References

Bastardie, F., Capowiez, Y., Cluzeau, D., 2005. 3D characterization of earthworm burrow systems in natural soil cores collected from a 12-year-oldpasture. *Appl. Soil Ecol.*, **30**: 34–46.

Capowiez, Y., Pierret, A., Daniel, O., Monestiez, P., Kretzschmar, A., 1998. 3D skeleton reconstructions of natural earthworm burrow systems using CAT scanimages of soil cores. *Biol. Fertil. Soils*, **27**: 51–59.

Curry, J., P., 1998. Factors affecting earthworm abundance in soils, In: *Earthworms Ecology*, Ed. C.A. Edwards, CRC Press LLC, Boca Raton, pp. 37–64.

Curry, J., P., 2004. Factors affecting the abundance of earthworms in soils. In: *Earthworm Ecology 2nd ed.*, Ed. C.A. Edwards, CRC Press, **91**: 114–1412.

Decaens, T., Bureau, F., Margerie, P., 2003. Earthworm communities in a wet agriculturallandscape of the Seine Valley (Upper Normandy, France). *Pedobiologia*, **47**: 479–489.

Easton, E. G., 1983. A guide to the valid names of Lumbricidae (Oligochaeta). In: *J.E. Satchell (ed.) Earthworm. Ecology – From Darwin to Vermiculture*. Chapman end Hall. London, pp. 475–487.

Graefe, U., Beylich, A., 2003. Critical values of soil acidification for annelid species and the decomposer community. *Newsletter Enchytraeidae*, **8**: 51–55.

Holdridge, L. R., 1947. Determination of world plant formations from simple climatic data. *Science*, **105**: 367–368.

Ivask, M., Kuu, A., Truu, M., Truu, J., 2006. The effect of soil type and soil moisture on earthworm communities. *J. Agricultural Sci*., **17**: 7-11.

Knapp, A. K., Fay P. A., Blair J. M., Collins S. L., Smith M. D., Carlisle J. D., Harper C. W., Danner B. T., Lett M. S., McCarron J. K., 2002. Rainfall variability, carbon cycling, and plant species diversity in a mesic grassland. *Science*, **298**: 2205–2202.

Knapp, A. K., Smith, M. D., 2001. Variation among biomes in temporal dynamics of above ground primary production. *Science*, **291**: 481–484

Kooch, Y. and Jalivand, H., 2008. Earthworms as ecosystem engineers and the most important detritivors in forest soil. *Pakistani Journal of Biological Sciences*, **11** (6): 819-825.

Kretzschmar, A., 1982. The burrow system of earthworms in grassland – seasonal variations of field observations. *Rev. Ecol. Biol. Sol.*, **19**: 579–591.

Lee, K. E., 1985. *Earthworms: Their ecology and relationships with soils and land use*, Acad. Press, Sydney, 411 pp.

Lieth, H., Whittaker R. H., 1975. Primary productivity of the biosphere. Springer, Berlin Heidelberg New York.

Malley, C., Nair, J., and Goen, Ho., 2006. Impact of heavy metals on enzymatic activity of substratum and on composting worms *Eiseniafetida. Bioresource Technology*, **97**:1498–1502.

Nordstrom, S., Rundgren S., 1974. Environmental factors and lumbricid associations in southern Sweden. *Pedobiologia*, **14**: 1–27.

Paoletti, M. G., 2001. *Invertebrate biodiversity as bioindicators of sustainable landscape*, Elsevier, Amsterdam, 446 pp.

Pop, V., 1949. Lumbricidele din România. *Anal. Acad. Rep.Pop. Române*, **1** (9): 383-505.

Rombke, J., Dreher, P., Beck, L., Hammel, W., Hund, K., Knoche, H., Kordel, W., Kratz, W., Moser, T., Pieper, S., Ruf, A., Spelda, J., Woas, S., 2000. Bodenbiologische Bodengute-Klassen – Umweltbundesamt Texte **600**: 1–275.

Sala, O. E., Parton, W. J., Joyce, L. A., Lauenroth, W. K., 1988. Primary production of the central grassland region of the United States. *Ecology*, **69**: 40–45.

Satchell, J. E., 1983. Earthworm ecology in forest soils. In: *Earthworm Ecology,* Ed. Satchell J.E. Chapman and Hall, London, 161–170.

Sommer, M., Ehrmann, O., Friedel, O., Martin, J. K., Vollmer, T., Turian, G., 2002. Boden als Lebensraum fur Organismen – Regenwurmer, Gehauselandschnecken und Bodenmikroorganismen in Waldern Baden-Wurttembergs – Hohenheimer Bodenkundliche **Hefte 63**: 163 p.

Whalen, J. K., Costa, C., 2003. Linking spatio-temporal dynamics of earthworm populations to nutrient cycling in temperate agricultural and forest ecosystems. *Pedobiologia*, **47**: 801–806.

Whittaker, R. H., 1975. *Communities and ecosystems*. MacMillan, New York.

EFFECTS OF SIMULATED ACID RAIN ON *IN VITRO* POLLEN PERFORMANCE OF *PYRUS COMMUNİS* L. AND *MALUS SYLVESTRİS* MİLLER

AYKUT TOPDEMİR and NAZMİ GUR
Firat University, Science Faculty Biology Department, 23169, Elazig, Turkey,
e-mail: atopdemir@gmail.com; nazmigur@gmail.com

Abstract. Successful fruit set depends on several reproductive processes including pollen germination and tube growth processes. Pollens of *Pyrus communis* L. and *Malus sylvestris* Miller flowers have been exposed to different concentrations of simulated acid rain solutions in range of pHs 6.0 to 2.8. Brawbaker Kwack medium was used for pollen germination. According to the results obtained, germination rate and pollen tube length for each plant at least has been affected at pH 6.0. Pollen germination and tube length significantly decreased with increasing acidity. The decrease of these rates for *Malus sylvestris* Miller was 99.44% for pollen germination and 99.68% for tube length; and for *Pyrus communis* L., 96.23% for pollen germination and 96.75% for tube length at a pH of 2.8. It has been shown that acidity has a higher effect on blocking *Malus sylvestris* Miller pollen performance *in vitro* comparative with *Pyrus communis* L.

Key words: Acid rain, apple, pear, pollen, germination, tube growth

Rezumat. Efectele ploii acide simulate asupra stării sporilor de polen de *Pyrus communis* şi *Malus sylvestris* Miller. Succesul fructificării depinde de o serie de procese reproductive ce includ procesele de germinare şi creştere a tubului polinic. Sporii de polen aparţinând speciilor *Pyrus communis* L. şi *Malus sylvestris* Miller au fost expuşi la diferite concentraţii ale unor soluţii de ploi acide simulate, cu pH-ul variind între 6,0 şi 2,8. Pentru germinarea polenului a fost folosit un mediu Brawbaker Kwack. Conform rezultatelor obţinute, rata de germinare şi lungimea tubului polinic au fost cel mai puţin afectate la un pH de 6,0. Atât rata de germinare a polenului, cât şi lungimea tubului polinic s-au micşorat considerabil odată cu creşterea acidităţii. Scăderea acestor rate la *Malus sylvestris* Miller au fost de 99,44% in ceea ce priveşte germinarea polenului şi de 99,68% pentru lungimea tubului polinic, iar pentru *Pyrus communis* L. a fost de 96,23% pentru germinaţia polenului şi de 96,75% pentru lungimea tubului polinic la un pH de 2,8. Aceasta a demonstrat că aciditatea are un efect mult mai inhibitor asupra stării polenului *in vitro* la *Malus sylvestris* Miller comparativ cu *Pyrus communis* L.

Cuvinte cheie: ploaie acidă, măr, păr, polen, germinaţie, tub polinic

Introduction

Acid rain is a serious environmental problem in the world. Studies investigating the effects of acid rain on the vegetative parts of plants, labeled these effects as biochemical (relationships between enzymes, proteins, amino acids, respiration and transpiration), microscopic (stoma destruction, epidermis and changes on the mesophyll layer) and macroscopic (necrosis, chlorosis and growth retardation) (Wolters and Martens, 1987; Shumejko *et al.*, 1996; Youngmi *et al.*, 2006; Sant'Anna-Santos *et al.*, 2006; He *et al.*, 2011). Acid rain is also effective on the generative parts of plants in addition to the vegetative parts. Pollen with male gametophytes among flowering plants is the principal part in this regard (Murdy, 1979). Pollen, reported as a bio-indicator of environment pollution by many researchers, is greatly affected by negative environmental factors (Wolters and Martens, 1987; Larrival *et. al.*, 1996). Acid rain is one of the factors that negatively

affect in vitro pollen germination and tube growth (Sidhu, 1983; Ryn *et al.*, 1986; Bellani *et al.*, 1987; Munzuroglu *et al.*, 2003).

This study aims to determine the effects of simulated acid rain on pollen germination and tube lenght, and thus establish the negative effects of acid rain on the pollen growth of apple and pear plants, which are of huge economic value for Turkey.

Materials and methods

Study material included the pollen of pear (*Pyrus communis* L.) and apple (*Malus sylvestris* Miller) plants in Elazığ Province and the surrounding counties. Brewbaker Kwack culture was used as a medium for the germination of the pollen (Shivanna and Rangaswamy, 1992). H_2SO_4 (Merck) and deionized water was used for preparing simulated acid rain solutions. Deionised water (pH 6.5) has been used as control.

pH concentrations were pH 6.0, pH 5.8, pH 5.6, pH 5.4, pH 5.2, pH 5.0, pH 4.8, pH 4.6, pH 4.4, pH 4.2, pH 4.0, pH 3.8, pH 3.6, pH 3.4, pH 3.2, pH 3.0, and pH 2.8. pH measurements have been done with digital pH meter (Extech PH100). Sterile 3 micro-slides were prepared for each pH solution (2 for experimental group, 1 for control group). A 50 µl culture solution was dripped to 2 various areas on each slide. Then 50 µl pH solutions (experimental groups) and 50 µl deionized water (control group, CG) were added onto the slides. Pollens on anther were homogeneously cultivated in the culture medium using a sterile syringe under a stereomicroscope. Petri dishes (15 cm diameter) with a moist filter paper lining the lower plate served as an improvised humidity chamber. Two glass rods were placed in parallel, about 4 cm apart, on the moist filter paper to facilitate the handling of the pollen cultures.

Then, the Petri dishes were settled in the incubator (Heraus B12) at 22 ±2 ^{o}C. Each germination medium was fixed with 10% ethanol after 3 hours to close the lamella. Germination percentages and tube lengths of the pollens were determined under the light microscope (Olympus, BX51) by the method of Shivanna and Rangaswamy (1992). All experiments were repeated three times and results were statistically analyzed by calculating variance and the standart error (Sx) of the mean. Statistical analysis was performed based on SPSS (version 10.0) program. In order to detect the significance of differencies ($p \square 0.01$ or $p \square 0.05$) of variables, a multiple comparison (LSD) test was performed.

Results

The effects of acidity on *in vitro* germination and tube lengths in the pollen of apple (*Malus sylvestris* Miller) and pear (*Pyrus communis* L.) plants are given in Figs. 1-4.

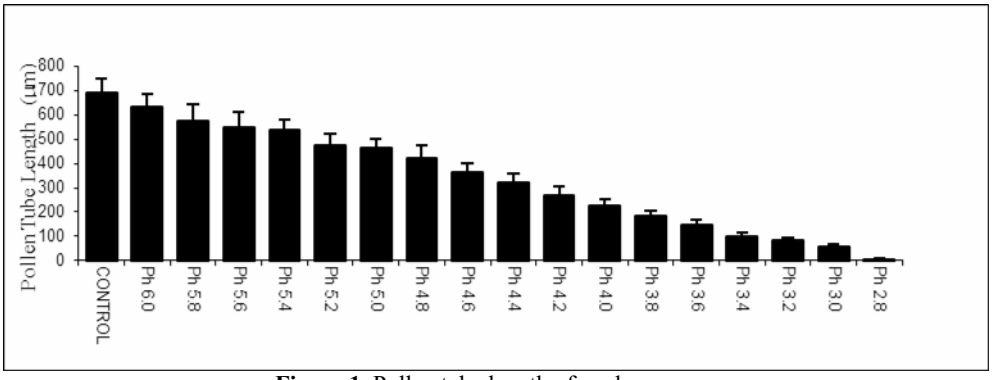


Figure 1. Pollen tube length of apple.

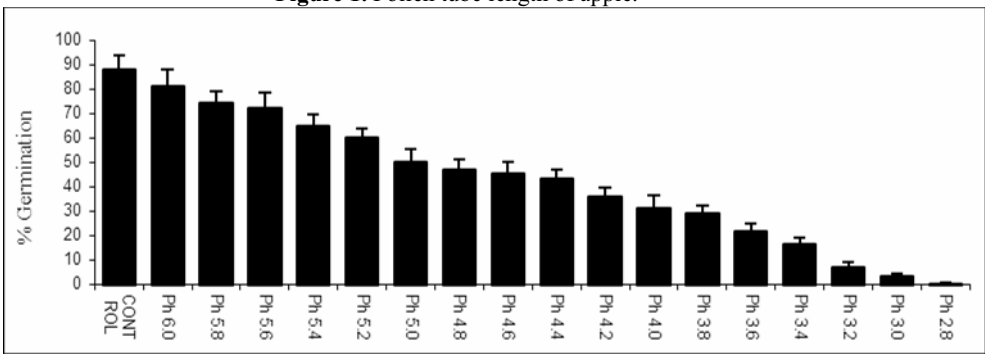


Figure 2. Pollen germination rates of apple.

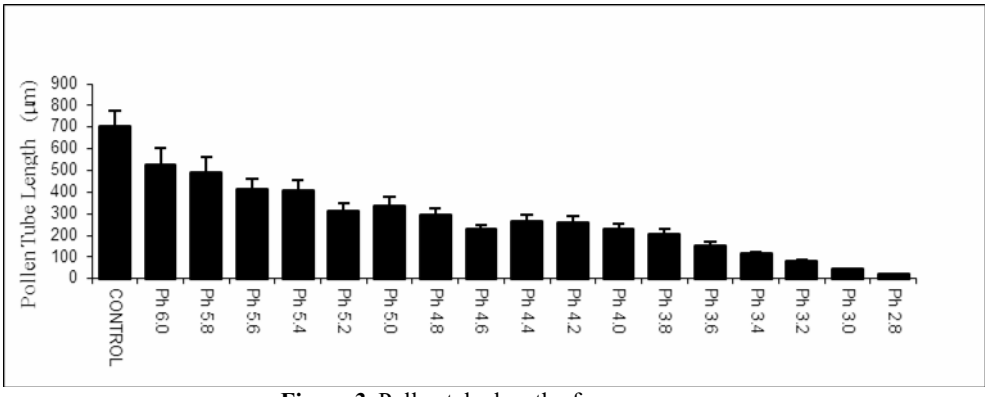


Figure 3. Pollen tube length of pear.

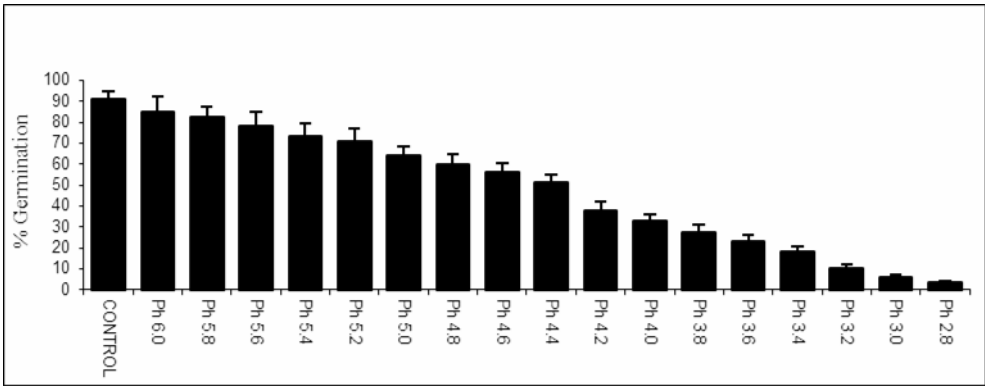


Figure 4. Pollen germination rates of pear.

The germination rate of the pollen of apple plant was determined as 88.2% in the control group (pH 6.5). The closest pH value (pH 6.0) to the control group was measured as 81.54% germination rate, and no significant ($p \square 0.05$) decrease was observed in germination rate. However, a regular decrease was determined in germination rate as the acidity degree increased. Germination rate was found to be negligibly low (0.49%) at pH 2.8. The comparison of the control groups and the highest and lowest acidity degrees indicated that 99.44% of decrease was observed at pH 2.8, and 7.57% of decrease was determined at pH 6.0.

In terms of the tube length in the apple plant, it was measured as 692 μm in the control group, and the difference was not significant at pH 6.0 and pH 5.8, the closest values to control group; however, the tube length significantly decreased with increasing acidity degree. These values are 81.2 μm at pH 3.2, 55 μm at pH 3.0, and 2.2 μm at pH 2.8. Considering the tube length of pollen, a comparison of the control group and tube length at pH 2.8 revealed 99.68% of decrease and statistically important ($p \square 0.01$) and the comparison with pH 6.0 indicated a 2.02 % decrease.

Pollen of the apple plant was negatively affected by high acidity both in terms of germination and tube length. The best germination and tube growth were observed in pH 6.0, closest to the control group.

The germination rates of the pollen of the pear plant indicated that there was 91.19% germination in the control group (pH 6.5). Depending on the increase in acidity, the germination rate was observed to decrease. This rate was 85.14% at pH 6.0 - the closest to the control group, and 82.34% at pH 5.8. These were not statistically significant ($p \square 0.05$). The germination rate was 6.34% at pH 3.0, and when it was compared to the control group, there was a 93.04% decrease in germination rate, and when 3.43% of the germination rate measured at pH 2.8 was compared to the control group, there was a 96.23% decrease ($p \square 0.01$).

The investigation of pear plant in terms of tube length demonstrated that tube length was 702.9 μm in the control group (pH 6.5), 632.4 μm at pH 6.0, the closest and lowest pH to the control group, and 22.8 μm at pH 2.8, the furthest and highest acidity value from pH control group. A comparison of tube length with the control group revealed a 10.01% decrease at pH 6.0, and a 96.75 % decrease at pH

2.8. As can be understood from these measurements, increasing acidity degree mostly inhibited the pollen tube length in the pear plant.

The comparison of inhibition rates in both plants demonstrated 96.23% in the pear plant and 99.44% in the apple plant at pH 2.8, and thus the apple pollen was concluded to be more affected. A comparison of pollen tube lengths showed that the apple plant was more affected by high acidity with a 99.68% decrease compared to pear plant.

Discussion

Acid rain is among the most important environmental factors affecting plant reproductive processes such as pollen germination, pollen tube growth and fruit-set. The current study, which examined pollen response to simulated acid rain from pH 6.0 to 2.8, shows very clearly that pollen tube germination and tube growth processes negatively effected from simulated acid rain.

Various studies have reported this subject for pollens of different plant species (Nadaka et al., 1980; Wertheim and Cracker, 1988; Renzoni and Viegi, 1991; Rinallo, 1992; Shan *et al.*, 1996; Sharma *et al.*, 1993).

In the study of Bellani *et al.*, apple pollen was subjected to acidity degrees at pH 5.6, 4.0 and 3.0. Apple pollen was examined for liveliness, germination and tube length. In parallel with the present study, pollen germination and tube length were observed to significantly decrease with increasing acidity degree (Bellani *et al*., 1987). Munzuroglu *et al.* (2003) obtained similar results in *Malus sylvestris* Miller cv. Golden pollen.

The present study demonstrates that simulated acid rain negatively affected the pollen germination and tube growth of economically important *Malus sylvestris* Millerand *Pyrus communis* L. plants. *Malus sylvestris* Miller is more sensitive to simulated acid rain than *Pyrus communis* L. In conclusion, considering the level of the exiting pollutant load of rain in the air, there is a serious cause of concern that pollen performance will be greatly reduced, with negative effects on apple and pear production, if the acidity of rainfall continues to rise.

References

Bellani, L. M., Rinallo, C., Muccifora, S., Gori, F., 1987. Effects of simulated acid rain on polen physiology and ultrastructure in the apple. *Env. Pol*. **95** (3): 357-362.

He, M., De-ming, D., Ju, W., 2011. Effects of simulated acid rain on main nutritional indicators of three leafy vegetables. *Chem. Res. in Chin. Univ.* **27** (3): 397-401.

Larrival, C., Bocquel C., Jarreau, M. C. C., Verhille, A. M., 1996. Polen: bio-indicator of pollution. J. of Aer. Sci. **27** (8): 227-228.

Munzuroglu, O., Obek, E., Geckil, H., 2003. Effects of simulated acid rain on the pollen germination and pollen tube growth of apple (*Malus sylvestris* Miller *cv.* Golden). *Acta Biol. Hun*. **54** (1): 95-103.

Murdy, W. H., 1979. Effect of SO_2 on sexual reproduction in *Lepidium virginicum* L. Originating from regions with different SO_2 concentrations. *Bot. Gaz*. **140**: 299-303.

Nakada, M., Fukaya K., Takeshita, S., Wada, Y., 1980. Effects of inorganic components in acid rain on tube elongation of camellia polen., *Environ. Pol.* **21** (1): 51-57.

Renzoni, G. C., Viegi, L., 1991. In vitro sensitivity *Pinus pinaster* and *P.pinea* pollen grains to different pH values. *Ann. Bot. Fenn.* **28**: 135-142.

Rinallo, C., 1992. Effects of simulated acid rain on the foliage and fruit yield of *Malus domestica* Borkh. *J. Hort. Sci.* **67**: 553-559.

Ryn, V. D. M., Jacobsen, J. S., Lassoie, J. P., 1986. Effects of acidity on the in vitro pollen germination and tube elongation in four hardwood species. *Can. J. For. Res*. **16**: 397-400.

Sant'Anna-Santos, B. F., Silva, L. C., Azevedo, A. A., Araujo, M., Alves, E. F., Silva, E. A. M., Aguiar, R. 2006. Effects of simulated acid rain on the foliar micromorphology and anatomy of tree tropical species. *Env. and Exp. Bot.* **58** (1): 158-168.

Shan, Y., Feng, Z., Izuta, T., Aoki, M., Totsuka, T., 1996. The individual and combined effects of ozone and simulated acid rain on growth, gas exchange rate and water-use efficiency of *Pinus armandi* Franch. *Environ. Pol*. **91**: 335-361.

Sharma, B., Singh, O. S., Khattra, S., 1993. Effect of acid rain on pollen germination and pollen tube growth in groundnut (*Arachis hypogea* L). *Indian J. Ecol*. **20**: 117-120.

Shivanna, K. R., Rangaswamy, N. S., 1992. *Polen biology laboratory manual*, Springer Verlag, New York, USA.

Shumejko, P., Ossipov, V., Neuvonen, S., 1996. The effect of simulated acid rain on the biochemical composition of Scots pine (*Pinus sylvestris* L.) needless. *Environ. Pol*. **92** (3): 315-321.

Sidhu, S. S., 1983. Effects of simulated acid rain on pollen germination and pollen tube growth of White spruce. Can. J. Of Bot. **61** (12): 3095-3099.

Wertheim, F. S., Craker, L. E., 1988. Effects of acid rain on corn silks and pollen germination. J. Env. Qual.17:135-138.

Wolters, J. H. B., Martens, M. J. M., 1987. Effects of air pollutants on pollen. *The Bot. Rev*. **53** (3): 372-414.

Youngmi, L., Park, J., Im, K., Kim, K., Lee, K., Park, J. A., Lee, T. K., Park, D. S., Yang, J. S., Kim, D., Lee, S., 2006. *Arabidopsis* leaf necrosis caused by simulated acid rain is related to the salicylic acid signaling pathway. *Plant Phys. And Bio.* **44** (1): 38-42.

INFLUENCE OF ADMINISTRATION OF DIFFERENT CHEMICAL FERTILIZERS AND BIOFERTILIZERS ON THE DYNAMICS OF HETEROTROPHIC BACTERIA, IN CHERNOZEM SOIL OF DOBROGEA

ELENA DELCA[1] and TATIANA PASCU[2]
[1]Ovidius University of Constanta, Mamaia Avenue, no. 124, 900552, Constanta, Romania, e-mail: delca_elena@yahoo.com;
[2]National Institute of Research and Development in Soil Science, Agrochemistry and Environment – ICPA Bucharest, Bd Marasti, no 61, 71331, Bucharest, Romania, e-mail: tpascu@yahoo.com

Abstract. The paper presents preliminary data on the effect of administration of inorganic or organic amendments upon the abundance dynamics of cultivated heterotrophic bacteria. Based on 7 experimental variants of cultivation was found that the total number of heterotrophic bacteria ranged between: $3.8x10^6$ CFU g^{-1} soil in April on the V_4 and $501.79x10^6$ CFU g^{-1} soil in May on the V_3, followed by V_6 in May, $142.13x10^6$ CFU g^{-1} soil. When soil was supplemented with manure, microbial density showed a significant increase $501.79x10^6$ CFU g^{-1} dry soil compared with control variant. Bacterial density increased significantly as value, too, following the administration of specific biofertilizers (Biovin, Bactofil Professional; Green Mycos).Inorganic fertilizers did not have a positive effect on microbial density values, being more or less similar to those reported for the control.

Key words: heterotrophic bacteria, soil fertility, biofertilizers, humus

Rezumat. Influența diferitelor îngrășăminte chimice și biofertilizatori asupra dinamicii bacteriilor heterotrofe cultivabile, în solul cernoziomic din Dobrogea. În lucrare se prezintă date preliminare privind efectul administrării unor amendamente organice sau anorganice asupra dinamicii abundenței bacteriilor heterotrofe cultivabile. Pe cele șapte variante experimentale s-a constatat că numărul total de bacterii heterotrofe cultivabile a variat între: minim $3.8x10^6$ UFC g^{-1} sol în luna aprilie pe V_4, și maxim $501.79x10^6$ UFC g^{-1} sol, în luna mai pe V_3, aceasta, fiind urmată de V_6 în luna mai, $142.13x10^6$ UFC g^{-1} sol. În cazul administrarii gunoiului de grajd densitatea microbiană a avut o creștere semnificativă $501.79x10^6$ UFC g^{-1} sol, față de varianta martor. Densitatea bacteriană a crescut la valori considerabile, deasemenea, în urma administrarii unor biofertilizatori specifici (Biovin, Bactofil Professional; Mycos Verde). Fertilizatorii chimici au avut un efect negativ asupra densității microbiene, valorile fiind mai mult sau mai puțin apropiate de cele înregistrate în cazul variantei martor.

Cuvinte cheie: bacterii heterotrofe, fertilitate sol, biofertilizatori, humus

Introduction

The research was conducted in the Turn of the Dobrogea, aiming to establish a pattern of chernozem soil biological reconstruction by applying various amendments aimed at stimulating bacterial abundance, with the targets:

- degradation and decomposition of organic matter
- restoration of soil structure
- recovery of the stock of humus.

Materials and methods

Experimental plot: 7 hectares of arable land outside the village Cumpăna, Constanta; Culture: Josef wheat.

Time of experimentation: the agricultural year 2009 - 2010 both in different phases of the wheat growing season and after harvest experimental versions: 7 plots.

Table 1. Fertilizers administered.

Experimental plots	Fertilizers administered
V1	100 kg.ha^{-1} $N_{15}P_{25}K_{15}$ in autumn, 150 kg.ha^{-1} NH_4NO_3 at the beginning of spring;
V2	Biovin 400 kg.ha^{-1}; Biovin 30 l.ha^{-1} - ½ at herbicide stage - ½ at flour stage;
V3	Garden soil – 15 t.ha^{-1} in autumn
V4	Biovin 30 l.ha^{-1} - ½ at herbicide stage; - ½ at flour stage;
V5	Biovin 150 kg.ha^{-1}- administered during sowing, NH_4NO_3 – 150 kg.ha^{-1} -40 kg.ha^{-1} at the beginning of spring; 50 kg.ha^{-1} at herbicide stage; 60 kg.ha^{-1} at flour stage.
V6	Biovin 375 kg.ha^{-1}; Biovin 30 l.ha^{-1} - ½ at herbicide stage; - ½ at flour stage; 1 mc Green Mycos, 1l Bactofil Professional;
V7	March – were not applied amendments.

Biovin Fertilizers are being administered for the first time in Dobrogea.

Biovin is being produced through a technological process from grape kernels. 12 years of research proved the following: it aerates the soil, improves it (it contains up to 70% humus makers- $8x10^7$ CFU/g), it enriches the soil with microorganisms that create humus ($8x10^9$ aerobic microorganisms per gram) (Berca, 2008).

Bactofil Professional is a product for improving the soil biological quality and contains nitrogen fixing bacteria $5.2X10^9$ CFU/ml, phosphate-solubilization bacteria, and heterotrophic bacteria that stimulate the decomposition of organic matter (Robescu, 2009).

Green Mycos is a product containing arbuscular mycorrhizal fungi and a number of factors that stimulate the establishment of symbiosis, is improving the soil quality up to 20 years (Berca, 2008).

The experiments have taken place on a 7 ha, which were divided in 7 variants, each variant being administered a different type of fertilizer in different quantities and periods.

The soil sampled was at a depth of 15 cm approximately, to perform a quantitative analysis of heterotrophic bacteria during the agricultural year 2009 - 2010 both in different phases of the wheat growing season and after watching his collection for development of microbial growth, depending on the variant.

Quantitative determination of microbial abundance was done by decimal dilutions of soil followed by inoculation of known quantities on solid nutrient media. (Clark, 1965). For this purpose, after weighing, the samples were inoculated on culture medium with a specific composition. Thus, to determine the number of total culturable heterotrophic bacteria it has been used nutrient:

- agar medium (Bergey's, 1986) - (pulvis yeast extract 2.5 g, peptone 0.2 g, Agar 17-20g. It was sterilized 20 min at 120^{o}C). There were inoculated Petri plates on each variant.

The total number of bacteria per gram of soil was calculated using the formula (Voiculescu *et al.*, 2006):

$$No.bacteria/g\ soil = \frac{no.colonies \times dilution \times 10 \times 100}{100 - U}$$

where:
X - average of colonies grown on culture medium,
10 - balancing coefficient of 0.1 ml of inoculum in the reporting of dilution soil
U% = soil moisture

Results and Discussions

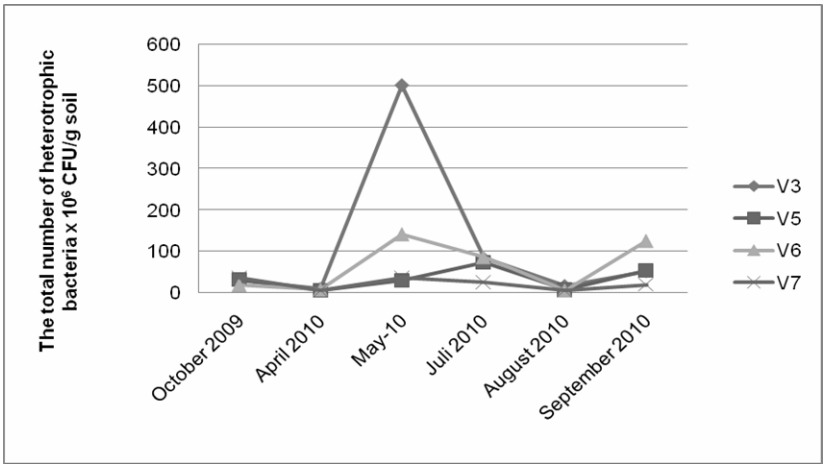


Figure 1. Dynamic variation of monthly heterotrophic bacteria in the crop year 2009-2010.

- Highest in May, the V_3 501.79x10^6CFU/g dry soil (Fig. 1), followed by the V_6 142.13x10^6CFU g^{-1} dry soil (Fig. 1) because fertilizer management, and favorable climatic factors:
  - moisture in the soil to a depth of 10 cm (16.1%), being 41.11 m^{-2} precipitation;
  - monthly average soil temperature at a depth of 10 cm (20.5ºC).
- V_5 present insignificant values close to those of the control variant.
- Major decline in August reflected in all variants (Fig. 1) as a result of unfavorable climatic factors:
  - low humidity (5.5);
  - monthly average soil temperature at a depth of 10 cm (31.3°C). (Data from the Research Station for Agricultural Development Valu Traian);
- Dynamics of heterotrophic bacteria in autumn 2010 shows a significant increase from the fall of 2009, the highest values being found in the V_6 125.93x10^6 CFU g^{-1} dry soil (Fig 1).

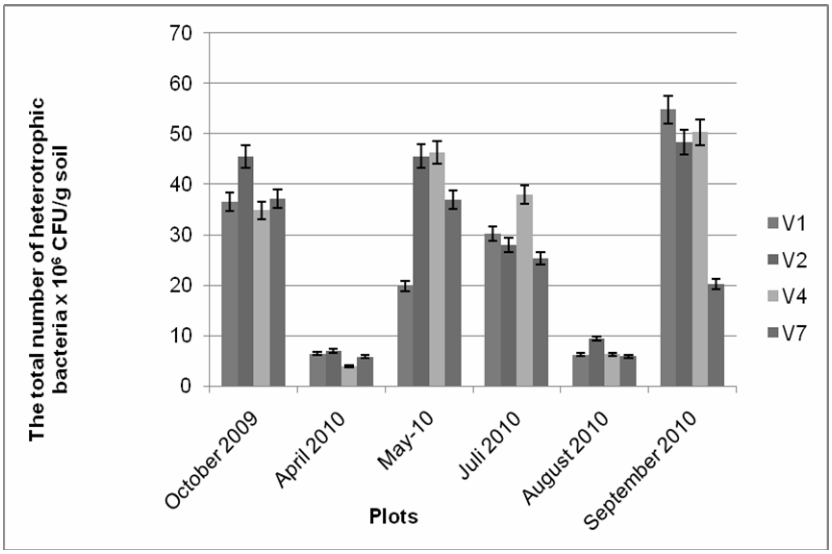


Figure 2. Dynamic variation of monthly heterotrophic bacteria in the crop year 2009-2010.

- V_1, V_2 and V_4 have insignificant values close to those of the control variant (Fig. 2).

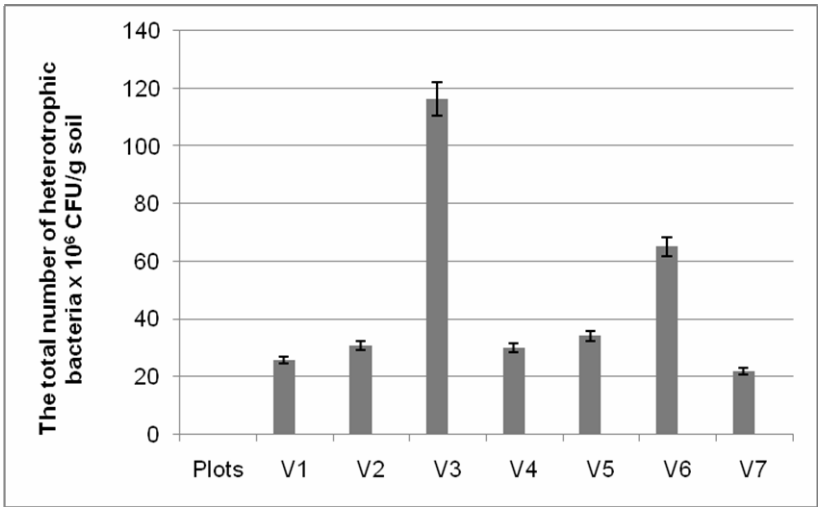


Figure 3. Annual average change in the dynamics of heterotrophic bacteria whichever.

Depending on the variant, the average annual cultivation of heterotrophic bacteria ranged from:

- maximum V_3 (manure) $116.32x10^6$ CFU g^{-1} dry soil (Fig. 3), followed by V_6 (Biovin, Bactofil Professional; Mycos Green) also showing a significant increase $64.99x10^6$ CFU g^{-1} dry soil (Fig. 3); minimum was found in V_1 $25.66x10^6$ CFU g^{-1} dry soil (Fig. 3) except for version control;
- V_1, V_2, V_4 and V_5 were recorded similar values, but significant upper control variant.

Conclusions

We recommend:

- V_3 (manure) as a model for biological reconstruction of the soil:
  - the highest abundance of heterotrophic bacteria $116.32x10^6$ CFU g^{-1} dry soil (Fig. 3)
  - show an increase of 531% compared to the control variant $21.89x10^6$ CFU g^{-1} dry soil (Fig. 3)
- V_6 (Biovin, Bactofil Professional; Green Mycos) has an abundance of heterotrophic bacteria $64.99x10^6$ CFU g^{-1} dry soil (Fig. 3);
  - show an increase of 358% compared to the control variant $21.89x10^6$ CFU g^{-1} dry soil (Fig. 3);

Microbial abundance values in the autumn of 2010 increased compared with the fall of 2009, except for control version. This increase in range can be explained by residual effect of biofertilizers apply, the amount of remaining organic matter in the form of plant debris after harvest.

Our preliminary data show that organic amendments with complex composition have a direct effect on the abundance and diversity of soil and influence indirectly the microbial metabolism and nutrient cycling rate.

References

Berca, M., 2008. *Probleme de ecologia solului*. Edit. Ceres, Bucuresti: 43-63.

Bergey's, 1986. *Manual of Sistematic Bacteriology*, vol. **2**, Williams and Wilkins, Baltimore, USA: 1075-1079.

Clark, F., 1965. Agar plate method for total microbial count. *Method for Soil Analysis*, vol. **2**: 1460-1465, Amercian Society for Agronomy, Madison, WL.

Robescu, V. O., 2009. *Modele privind managementul reconstructiei de mediu in bazinul superior al raului Dambovita*. Edit. Ceres, Bucuresti: 189-196.

Voiculescu, A. R., Toti, M., Dumitru, M., 2006. *Decontaminarea solurilor poluate cu compusi organici,* Ed. SITECH, Craiova

SOCIO-MEDICAL IMPLICATIONS OF SYPHILIS

VASILE SIRBU and IRINA-LILIANA FLOREA
"Alexandru Ioan Cuza" University of Iasi, B-dul Carol I, No. 20 A, 700505, Iasi, Romania,
e-mail: sirbu.vasile@email.ro

Abstract. This work is a study on the cases of syphilis diagnosed and recorded at the Dermatology Clinic of "Sf. Spiridon" Hospital of Iasi (2007-2009). The socio-medical implications of the disease were established monitoring the number of cases by: date of recording, gender, age groups, disease stage of evolution, environment of residence (urban or rural); the patients' marital status, occupation, the infection's way of transmission; the patients' tracking.

Key words: syphilis, socio-medical implications

Rezumat. **Implicații socio-medicale ale sifilisului.** Prezenta lucrare este un studiu asupra sifilisului. Materialul provine din cazurile înregistrate şi tratate la Clinica de Dermatologie, Spitalul "Sf. Spiridon", Iaşi (2007-2009). Implicațiile medico-sociale ale bolii au fost stabilite prin monitorizarea cazurilor depistate urmărind: data înregistrării pacientului, sexul, vârsta, stadiul de evoluție a bolii, mediul de reşedință (urban sau rural), statutul social, ocupația, modul de infectare şi urmărirea pacienților.

Cuvinte cheie: sifilis, implicații medico-sociale

Introduction

Sexually transmitted diseases are important through the repercussions they have over population health, being involved in both female and male infertility, in the etiology of abortion, mortality and prenathal pathology, in uro-genital pathology (Bădănoiu *et al.*, 1982; Bucur & Giurcăneanu, 2000).

In last decades, there is a slight but constant increase, 10-15% per year of the sexually transmitted diseases (Bucur & Opriş, 2002; Conu, 1976). On this trend, there are periods of one or two years with an increased number of cases. After OMS, every year over 100.000.000 cases are recorded, beeing along the moust numerous grup of transmited disease (Forsea *et al.*, 1996; Popescu & Longhin, 1969).

Before descovering HPV infections, syphilis was considered the moust dangerous sexual disease (Halioua & Lunel-Fabiani, 2003; Popescu, 1982).

Materials and methods

This work is a study on the cases of syphilis diagnosed and recorded at the Dermatology Clinic of "Sf. Spiridon" Hospital of Iasi (2007-2009).

The socio-medical implications of the disease were established monitoring the number of cases by: date of recording, gender, age groups, disease stage of evolution, environment of residence (urban or rural); the patients' marital status, occupation, the infection's way of transmission; the patients' tracking.

Results and discutions

From the distribution of the total number of patients, we can observe a decrease of the number of cases recorded in 2008 and 2009, compared to 2007, a fact which might indicate a better assimilation of the measures of individual

prophylaxis (use of condoms, avoiding random sexual intercourse, washing sexual organs using soup and water after sexual intercourse) and antivenereal sanitary education (way of transmission and early signs, major complications, advantages of corect and early tratement, the importance of knowing exact source of infection) (Table 1).

Table 1. Syphilis cases, by date of recording, 2007-2009.

Year	Number cases	%
2007	111	35.30
2008	106	33.60
2009	98	31.10
TOTAL	315	100.00

The number of cases is slightly higher in the male population (167 cases-53%), compared with female population (148 cases-47%), due to the ease of establishing random sexual intercourse, when practicing a profession that involves long separations from the family. Though the women are important sources of syphilitic infection spread. If between 2007-2008 the ratio male-female cases is increased between males, in 2009 the ratio is 41 male cases and 57 female cases (Table 2). Syphilis cases by age groups, indicated the highs value between 20-24 years (26.3%), followed by the 15-19 years age grup (18.1%), almost 50% of cases. From this reason, the sexual education must begun very early (Table 3). We can observe a higher number of patients in the urban environment 208 cases (66%), comparing with 107 cases (34%) in rural areas, which can be explained through the existence of groups with high syphilis risk, also taking into account that the rural environment imposes a family traditional environment (Table 4).

Considering the patients' marital status, the highest number of decays, amounting to 195 cases (62%), is recorded among the unmarried or separated persons, due to the predisposition to random intercourse and to the absence of a stable partner. The marital status confer a stable state, only 120 cases (38%) being recorded (Table 5). From the total of 315 cases, 191 cases are unemployment, among them being the most numerous cases, they easily have random intercourses.

Table 2. Syphilis cases, by gender, 2007-2009.

Gender	**Number of cases 2007**	**%**	**Number of cases 2008**	**%**	**Number of cases 2009**	**%**
Male	64	57.00	62	58.00	41	42.00
Female	47	43.00	44	42.00	57	58.00
TOTAL	**111**	**100.00**	**106**	**100.00**	**98**	**100.00**

Table 3. Syphilis cases, by age grups, 2007-2009.

Age grups	**Number of cases**	**%**
0 - 15 years	5	1.60
15 - 19 years	55	18.10
20 - 24 years	83	26.30
25 - 29 years	55	17.50
30 - 34 years	35	11.40
35 - 39 years	24	7.30
40 - 44 years	27	8.00
45 - 49 years	17	5.40
over 50 years	14	4.40
TOTAL	**315**	**100.00**

Table 4. Number of cases, urban and rural areas, 2007-2009.

Residence	**Number of cases**	**%**
Urban	208	66.03
Rural	107	33.97
TOTAL	**315**	**100.00**

Table 5. Syphilis cases, by marital status and gender, 2007-2009.

Marital status and gender	**Number of cases**	**%**	**Marital status and gender**	**Number of cases**	**%**
Married female	62	42.00	Married man	59	35.30
Unmarried female	86	58.00	Unmarried man	108	64.70
TOTAL	**148**	**100.00**	**TOTAL**	**167**	**100.00**

Concerning infection's way of transmission, the sexual behavior is important, in this way a great number of infectious being made by random intercourse 144 cases (44.4%). Some of the pacients where infected from friends 23 cases, 57 could not give information about the way of transmission of the disease.

The patients were tracked by the help of the state epidemiologic organizations (20%), by the usual medical control (28.6%), by medical control at the

working places (8.7%), prenatal medical examination (6.6%) and pre-marital medical examination (2.8%).

Conclusions

From the distribution of the total number of patients, we can observe a decrease of the number of cases recorded in 2008 and 2009, compared to 2007, a fact which might indicate a better assimilation of the measures of individual prophylaxis and antiveneral sanitary education.

The number of cases is slightly higher in the male population due to the ease of establishing random sexual intercourse, when practicing a profession that involves long separations from the family. Though women are important sources of syphilitic infection spread (Grigore & Țolea, [s.a.]).

We can observe a higher number of patients in the urban environment, which can be explained through the existence of groups with high syphilis risk, also taking into account that the rural environment imposes a family traditional environment.

References

Bădănoiu, A., Toma, D., Țolea, I., Talir, A., 1982. *Dermato-Venerologie*, Bucureşti.
Bucur, G., Giurcăneanu, C., 2000. *Boli transmise pe cale sexuală*, Editura Celsius, Bucureşti.
Bucur, G., Opriş, D. A., 2002. *Boli Dermatovenerice*, Enciclopedie, Editura Medicală Națională.
Conu, A., 1976. *Dermatovenerologia*, Editura Didactică şi Pedagogică, Bucureşti.
Forsea, D., Popescu, R., Popescu, C. M., 1996. *Compendiu de Dermatologie si Venerologie*, Ed. Tehnică, Bucureşti.
Grigore, T., Țolea, I., [s.a.] *Boli cu transmitere sexuală la femeie,* Editura INFO.
Halioua, B., Lunel-Fabiani, F., 2003. *Bolile cu transmitere sexuală*, Editura Corint, Bucureşti.
Oanță, A., *Curs de Dermatologie*, 2007. Editura Universitatii Brasov.
Popescu, A., 1982. *Boli cu transmisie sexuală*, Editura Medicală, Bucureşti.
Popescu, A., Longhin, S., 1969. *Biologia Treponemei pallidum*, Editura Academiei, Bucureşti.

THE ROLE OF ECO-EDUCATION IN PROMOTING ENVIRONMENTAL ETHICS

MIRCEA NICOARA
"Alexandru Ioan Cuza" University of Iași, Bd. Carol I, 20A, Iasi, 700505, Romania,
e-mail: mirmag@uaic.ro

"The purpose of environmental education is to help students become aware / sensitive to the environment, well informed, skilled, committed citizens employed to work individually and jointly, to protect, enhance and sustain the environment, from behalf of the present and future generations of all living organisms." (Wisconsin - Department of Public Instruction)

Abstract. The education does not resolve the conflicts between technological progress and economic and social development, and the need to protect the environment that provides the necessary resources. We can choose between the ways of our needs fulfillment, between a sustainable development with nature and resources conservation, and a wild one which degrades the environment. Environmental education must be holistic, lifelong, interdisciplinary, and in line with the rapid changes taking place, preparing the human individual for life. The citizens of a democratic society based on knowledge should make an informed choice between options related to responsible scientific and technical progress. Increasing technological risk and associated ethical issues raise questions and reinforce the public desire for progress to be closer supervised. The scientific and technological progress changes people's lives too fast, leading to serious ethical issues, with potential implications on future generations. In ecology, ethics must meet the new discoveries and new thinking, to protect the natural environment and human society. Knowledge gained through research must serve the humanity and nature; otherwise research should be halted in the name of an ethics for man and nature. In this thought entered the "neonaturalistic pantheism", "return to a new barbarism" and "environmental robinsonism". The return to a non-technical civilization based on traditional agriculture, crafts and home economy is a false solution to global ecological crisis. The solution to the contemporary ecological crisis is to dispense with the anthropocentric ethics and give moral consideration to the non-human beings. A different, more biocentrical paradigm is needed. The eco-conscience requires an unprecedented mixture between science and conscience, biology and ethics. The last challenge of environmental ethics is the preservation of life on Earth. A theory of the value is needed to support the new ethics and new model of conduct: a metaphysics related to land, a metaecology.

Key words. eco-education, environmental ethics, metaecology

Rezumat. Rolul eco-educației în promovarea eticii ambientale. Educația nu soluționează conflictele dintre continuarea progresului tehnologic și a dezvoltării economico-sociale, și necesitatea protejării mediului care ne oferă resursele necesare. Putem alege, însă, între modul de împlinire a nevoilor noastre, între o dezvoltare durabilă, cu conservarea naturii și a resurselor, și una sălbatică, care degradează și distruge mediul. Educația pentru mediu trebuie să fie holistă, pe tot parcursul vieții și interdisciplinară, și să fie în acord cu schimbările rapide ce au loc în prezent, pregătind individul uman pentru viață. Cetățenii unei societăți democratice, bazate pe cunoaștere, trebuie să fie capabili să facă o alegere informată între opțiunile legate de progresul tehnico-științific responsabil. Riscul tehnologic în creștere și temele etice asociate ridică întrebări și întăresc dorința publicului ca progresul să fie mai îndeaproape supravegheat. Progresul științific și tehnologic schimbă viețile oamenilor prea rapid, dând naștere la serioase probleme de etică, cu implicații potențiale asupra generațiilor viitoare. În ecologie, etica trebuie să corespundă noilor descoperiri și noului mod de gândire, pentru protecția mediului natural și, deci, a societății omenești. Această cunoaștere dobândită prin cercetare trebuie să servească umanitatea și natura, în caz contrar, cercetările să fie oprite în numele unei etici pentru om și natură. În această gândire se înscriu „panteismul neonaturalist", „întoarcerea la o nouă barbarie" și „robinsonismul ecologic". Întoarcerea la o civilizație atehnică, bazată pe agricultură tradițională, meșteșuguri și economie domestică este o falsă rezolvare a crizei ecologice globale. Soluția la criza ecologică contemporană constă în renunțarea la etica antropocentrică și considerarea morală a ființelor non-umane. Este necesară o paradigmă diferită, mai

biocentrică. Eco-conştiinţa necesită amestecul fără precedent între ştiinţă şi conştiinţă, biologie şi etică. Ultima provocare a eticii mediului constă în conservarea vieţii pe pământ. Este necesară o teorie a valorii care să sprijine noua etică şi noul model de conduită: o metafizică legată de pământ, o metaecologie.

Cuvinte cheie: eco-educaţie, etică ambientală, metaecologie

Many people in urban industrial countries are not aware about the environmental consequences of their actions. For many more people from developing countries there is not an alternative to their daily struggle for survival besides the consumption of any available resource, without taking into account the long-term environmental damages. Education cannot resolve conflicts between the immediate desperate needs for productive land, food and fuel, and the need to support environment which all these resources are depending on. However, there are viable choices between modes more or less acceptable for fulfillment of human urgent needs, between the development taking place with nature and resources conservation, and the one that degrades and destroys the environment.

The environmental conscience is a *sine qua non* component of the social conscience. Environmental education (eco-education), in addition to scientific (ecological, economic, medical, health, etc.) justification, offers the premises of a plenary life, a Man in harmony with nature and with himself. Man of ecological crisis suffers the drama of the conflict between values (e.g., the beauty of a natural landscape can be sacrificed to pragmatic usefulness). The education related to environment must be holistic and lifelong, and to reflect the changes in a universe in rapid transformation. It must be interdisciplinary and to prepare the human individual for life, giving him the understanding of the interdependence between natural and social environments.

A fifth of UNEP's budget is spent on staff education: more than 30,000 technicians, teachers, decision staff, etc. since 1973. The training interests are programs for: pest and vector control, resource conservation, regional seas, etc.

An international workshop on environmental education was held in Belgrade, in 1975. In the same year, UNESCO and UNEP launched the International Environmental Education Program (IEEP). The "World Conference on Environmental Education" held in Tbilisi (October 1977) established the guidelines for environmental protection studies promotion in schools and universities. The main purpose was to formulate recommendations for action at national, regional and international levels to promote and develop environmental education.

Four regional meetings of experts on education related to environment occurred in 1976:

- Brazzaville for Africa;
- Kuwait for Arab countries;
- Bogota for Latin America and the Caribbean;
- Helsinki for Europe.

In 1987, after Moscow Congress on environmental education and teaching, UNEP and UNESCO have published the international strategy on education and teaching for 1990, which currently serves as the basis of national strategies. IEEP, UNEP and UNESCO have trained about 12,000 environmental educators and

teachers. IEEP periodical "Connect" appears in 6 languages and is the most widespread publication of UNEP and UNESCO.

A new European initiative is to develop a stronger and more harmonious relationship between the world of science and the society in general. It outlines 38 actions related to the place of science in education and popular culture, involving citizens in science policy, gender dimension, ethics and governance. While the European Commission intends to act as a catalyst force, these actions will require concerted effort of the actors from the Member States, regions, groups of shareholders and interested citizens. The plan provides a common framework to monitor progress and evaluate results.

In a knowledge-based society, the democratic governance must ensure that citizens are able to make an informed choice between the options offered by responsible scientific and technological progress. Only half of the consulted Europeans declared they are interested in science and many of them consider themselves as poorly informed. Primary intention is to support the strategic objective set by the EU in Lisbon, to become the most competitive and dynamic knowledge-based economy in the world and, at the same time, is part of the process of creating the European Research Area, process launched by the European Commission in January 2000.

The conference "Science and Governance" took place in October 2000. Europeans' attitudes towards science (as the Eurobarometer survey of October 2001) ranged from trust and hope to lack of interest or even fear against the impacts of the scientific activities. The industrial risks and ethical issues are intensely mediated, raising questions and enhancing public desire for progress to be more closely monitored. Some people feel that science and technology change their lives too quickly. Although there has been progress, too many stereotypes still keep women out of science. Young people do not find the study of science and the scientific careers attractive enough. The labor market is affected; the industry is struggling to recruit engineers and scientists.

Environmental awareness: the number of European members of Greenpeace has doubled since 1988. Contributions to the World Wide Fund for Nature, former World Wildlife Fund, increased from 300,000 to 1.8 million pounds in 1992. One from five people questioned in Spain, Sweden, Germany, and Holland chose "care about environment" as the most sought after quality of a potential supplier.

In the field of Ecology, the general ethics must mould on the characteristics of the new discoveries and new thinking, for a bioethics useful to natural environment protection and therefore to human society. Social and behavioral sciences have not progressed at the same pace with the natural sciences, in particular with the biological ones, and the value systems not readjusted according to the structure of modern society. The genetic manipulation leads to production of less costly and more effective hormones and vaccines, to replacement of chemical fertilizers with nitrogen fixing symbiotic bacteria, to bacterial decomposition of oil and its derivatives. Handling hasty, without sufficient precautions, could result in release into the environment of manipulated organisms containing hybrid molecules able to induce aberrant genetic information and great damage in people, flora and fauna, once out of control. Species that did not exist on Earth could be produced.

Some even proposed that such research to be stopped in the name of an ethics for man and nature. Ethics must ensure that the knowledge gained through research serves humanity and nature.

The satisfaction felt in nature (even in the urban "green areas") may be an echo of primitive *Homo naturalis* in the present "artificialized" man. The alternative of return to an atechnical civilization based on traditional agriculture, crafts and home economy is a false solution to global environmental problems. Variants of this mentality are the "neonaturalistic pantheism", "return to a new barbarism", and "environmental robinsonism".

The environmental ethics claims that we can progress in ending the ecological crisis, if we change our anthropocentric ethical attitude and give moral consideration to the non-human beings. The holistic approach of environmental ethics requires a different theoretical paradigm, more biocentric. A reordering of our moral universe is needed. An ethics is a system of principles or a theory which includes a set of values and a set of overall evaluative judgments on conduct, what is compulsory, allowed or wrong, which are the rights, what has value and so on. "The balance of nature" is not itself a moral rule, but plays an important role in our overall image on the natural world, which includes the attitude of respect for nature. "The welfare of individual organisms considered as entities with inherent value must determine our moral relations with wildlife communities on Earth. The good for a non-human specimen would be the complete development of its biological powers." The welfare of a population or community of non-human individuals consists in maintaining them from generation to generation as a coherent system of genetic and environmental related bodies, whose average welfare is at an optimal level in relation to the given environment. An attitude is correct, if it tends to preserve the integrity, stability and beauty of the biotic community (by Aldo Leopold). We have a moral obligation to endangered species (e.g., conservation of whales). Conservation is a state of harmony between people and Earth. No important ethical change was ever done without an internal change in our intellectual accents, beliefs, emotions and convictions. The proof that conservation has not yet reached these foundations of conduct lies in the fact that philosophy and religion have not yet heard about it. In our attempt to make conservation easier, we turned it into a common place.

In the past, conservation concerned the species of economic value only. But most members of the biotic community have no economic value, and still help maintain it and are entitled to continuity. A subterfuge was reached: when a non-economic category is threatened, and it happens to be liked, an economic importance is invented (e.g., songbirds protect us from insects). The same occurred for predatory mammals, predatory and ichthyophagous birds. They would help to game quality maintaining by killing the weak individuals, rodent species harmful for farmers, or by feeding on "worthless" species. The evidence must be economic to be valid here as well. Only in recent years were heard more honest arguments about the predator place in community and that no special interest gives the right to exterminate them for the sake of a real or imagined benefit.

Some species of trees were removed from the calculation by foresters with economic thought (USA), because they grow too slowly or give too low value

timber (white cedar, American larch, cypress, beech). In Europe, where forestry is ecologically more advanced, a functional value in building soil fertility was found for the non-commercial species of trees. The interdependence between forest constituent species of trees, flora and fauna is recognized. The lack of economic value characterizes not only the species or groups, but entire biotic communities: marshes, peat bogs, dunes and "deserts". In some cases, the alleged lacks of profit of these „empty" areas proved wrong, but only after many were destroyed. Efforts to re-flood the peat bogs are made today.

Earth is not only soil, is a fountain of energy flowing through a circuit of soils, plants and animals. The food chains are living channels leading the energy upwards. The death and decomposition return it to the ground. The circuit is not closed, part of the energy dissipates by decay, some is added by air absorption, and part is stored in soils, peat and ancient forests.

An ethical relationship with the land cannot exist without love, respect and admiration for the land, and a high consideration for its value understood not in the economic sense, but in a much broader, philosophical sense. Perhaps the most serious obstacle to a land ethics is that our educational and economic system is not oriented, but deviated from an intense conscience of the land, being satisfied with surrogates instead of land natural products. Another obstacle is the attitude of farmers who still consider the land as an enemy or an owner keeping them in slavery.

The evolution of an ethics of Earth is both an intellectual and emotional process. As the ethical frontier advances from individual to community, its intellectual content increases. Operating mechanism is the same as for any ethics: social approval for corrective actions, social disapproval for wrong actions.

We, humans, are full members and citizens of the biotic community and we cannot make an exception by uncontrolled population growth. At the same time, however, we remain members of the human community or "global village", with correlative moral responsibilities. So, we are moral beings, not despite but in harmony with nature. The challenge of environmental ethics is an attempt to redefine the borders of ethical obligation, an invitation to moral development. A comprehensive ethics will find values in and debts to natural world. An environmental conscience requires unprecedented blend of science and conscience, biology and ethics.

An organism is "good" or "bad" only in terms of human interests. It becomes difficult to say whether one species is "bad" throughout the ecosystem. An "enemy" (predator) may be even good for the "victimized" species (prey) (e.g., predation maintains the herd healthy). One may only say that natural things are good until proven otherwise. A species has the right to exist. The species only exist in the form of plant or animal individuals. Biologically, the right to life is an adaptive disposition valid for life, which survives over millennia, and this leads to the assumption that species are good in their niches exactly and, therefore, is correct for people to leave them where they are and let them evolve.

Those wishing to restore the rare species of big cats in the wild, raised the problem of killing inferior specimens, genetically related, present in zoos, in order to accommodate individuals needed to rebuild and maintain a population genetically

able to survive after release. E.g., all Siberian tigers in North America zoos are descendants of seven individuals.

What people do evil or allow happening by negligence is to stop the historical vitality of life, the natural flow. Any extinction is a kind of super killing. It kills forms (species) instead of individuals. It kills "essences" besides "existences." It kills collectively, not just distributively. It kills the birth, as well as the death. One thing is correct (by Aldo Leopold), if tends to preserve the integrity and stability of the biotic community. Otherwise it is wrong. Hence two points: that ecosystems exist both in the wild and in support of culture, and that they must exist both for what represent in themselves and as modified by culture.

The environmental ethics must be geared to ecosystems. The concern should be the basic unit of survival. E.g., the restoration of the wolf population – top of the food pyramid – in order to restore the full integrity, stability and beauty of Yellowstone National Park, despite the sufferings of the preys (elks, etc.); as necessary as fire for plant succession in the forest.

The environmental ethics must pass the boundary set by some conservatives between the "real" organisms and "nominal" ecosystems. Unlike higher animals, the ecosystems have no experience; unlike plants, they have no organized centre - a genome, nor defend themselves against injury or death. There is no instrumental or intrinsic value for ecosystems. A third category - the systemic value - must be introduced.
Once, the environmental ethics said that ecosystems are valuable because of their contribution to animal experiences or to organismic life. Later, a deeper vision, more conservative and more radical, considered that the stability, integrity and beauty of biotic communities are to be fundamentally preserved.

In practice, the last challenge of the environmental ethics is life preservation on Earth. In principle, the last challenge is a theory of value, sufficiently deep to support this ethics. We must have a record of the values held by nature, and an ethics that appropriately respects those values. Such values equally belong to biology of the natural history and psychology of the human experience. Some of the nature values depend on us, but the beauties of Earth do not simply consist of their roles as resources for humans, support for culture or experience stimuli. There is no value without the assessor, but the natural system vitality or values are not in the human mind only. The assessment opportunity is undertaken for us by the evolutionary and ecological natural history, and such nature is already valuable, before people come to evaluate what happens. Some values are anthropogenic, created by people, but others are biogenic, with natural causes. In the environmental ethics, one's beliefs about nature, which are based on, but exceed science, are closely linked with beliefs about duty. The way the world is informs about how it must be. We shape our values to a significant extent in agreement with our concept about how the universe we live in is, and this leads our sense of duty. Our model of reality involves a pattern of conduct. For an environmental ethics, we need a metaphysics related to land, a metaecology.

References:

Commoner, B., 1970. *Ştiinţă şi supravieţuire*, Edit. Politică, Bucureşti.
Commoner, B., 1976. *Cercul care se închide*, Edit. Politică, Bucureşti.
Ionescu, A., Săhleanu, V., Bindiu, C., 1989. *Protecţia mediului înconjurător şi educaţia ecologică*, Edit. Tehn., Bucureşti.
Lang, S., 2000. *Integrarea europeană-un mediu mai curat? Raport asupra percepţiei publice cu privire la procesul de integrare europeană şi mediul înconjurător în România*, Centrul Regional de Protecţia Mediului pentru Europa Centrală şi de Est, Szentendre, Ungaria.
Mandravel, C., Guţul-Văluţă, M., 1987. *Pământenii-duşmanii Terrei?*, Edit. Albatros, Bucureşti.
McKinney, M. L., Schoch, R., 1998. *Environmental Science*, Jones and Bartlett Publishers Inc., London.
Mohan, G., Ardelean, A., 1993. *Ecologie şi protecţia mediului înconjurător,* Edit. „Scaiul", Bucureşti.
Negulescu, M., Ianculescu, S, Vaicum, L., Pătru, C., Bonciu, G., 1995. *Protecţia mediului înconjurător*, Edit. Tehn., Bucureşti.
Nicoară, M., 2009. *Legislaţie, instituţii şi politici de mediu*, Editura Tehnopress, Iaşi.
Nicolaescu, G., 2002. *Educaţie pentru mediu*, Edit. Carminis, Piteşti.
Pumnea, C., Grigoriu, G, 1994. *Protecţia mediului ambiant,* Edit. Did. şi Ped. R.A., Bucureşti.
Vişan, S., Creţu, S., Alpopi, C., 1998. *Mediul înconjurător. Poluare şi protecţie*, Edit. Econ., Bucureşti.
Zimmerman, M., 1993. *Environmental Philosophy: from animal rights to medical ecology*, Prentice Hall, Englewood Cliffs, New Jersey.
*** 1997. *The European Union and the Environment*, Luxembourg, Office for Official Publications of the European Communities.
*** 1999. *A future for Europe's nature and biodiversity*, Proceedings, European Centre for Nature Conservation – Tilburg.
*** 2000. *Environment for Europeans*, No. 5, European Commision.
*** 2002. *Choices for a greener future, The E.U. and the environment*, Office for official publications of the European communities, Luxembourg.

FEW MEMORIES CONCERNING THE MARINE BIOLOGICAL STATION "PROFESSOR IOAN BORCEA" FROM AGIGEA–CONSTANTA AT THE 85-TH ANNIVERSARY

CONSTANTIN TOMA
"Alexandru Ioan Cuza" University of Iasi, Faculty of Biology, Bd. Carol I, 20A, 700505, Iasi, Romania

About the station from Agigea I heard in 1953 only, when I became a student of Faculty of Natural Sciences from "Alexandru Ioan Cuza" University from Iaşi. At that time, I learned that the first modern university had the oldest Museum of Natural History (founded in 1834) and the oldest Botanical Garden (founded in 1856). I was proud about all these, as well as the gold gallery of ours professors from whom I lectured courses, seminaries and practical training stages.

I was extremely happy when I saw that, at the end of second academic year, practical training stage will be held in the Black Sea areas at the station of our university. I knew something about the Black Sea from my teachers of Geography in high-school. I could not wait to see it. And surely that day came! The Agigea Resort, in mid June of 1955, we have been waited by Station Director himself (Professor Sergiu Cărăuşu). He was the professor who taught us, in the fourth academic year, about Hydrobiology domain. At that time, naturalist-students performed one entire month of practical training stage at the end of each of first four years of study (with all costs provided by the university).

Once inside of Agigea Resort, Director Sergiu Cărăuşu together with our teachers (Professor Mihai Constantineanu, Lecturer Paul Borcea and Assistant Napoleon Topală) presented laboratories, museum, the famous reservation areas with its famous dunes and the specialized library. After that, we met the Romanian researchers who worked inside of this sanctuary of biology. I still remember PhD Aurelia Cărăuşu, Maria Celan and Porumb family well-known and appreciated zoologists, hydrobiologists and algologists. I cannot forget, even after 55 years, the ambient of these laboratories in which they worked: the well-equipped laboratories for those times. There, they made researches concerning the environment, seaside and Black Sea waters. They made observations, collecting invertebrate animals and algae. During our stage, they learned us to process and preserve all kind of collected samples using - awkwardly in the beginning - determination keys of flora and fauna, making some simple experiments with jellyfish and molluscs.

After the first two academic years of study completed, I had sufficient knowledge of botany and zoology, parasitology and entomology, physics and chemistry, which allowe me to understand biological diversity of Black Sea and its depths. As a member of Vertebrate Zoology scientific circle, led by Professor Zicman Feider, my scientific interest was focused on ichthyology (I was fascinated by a treatise published by Professor Sergiu Cărăuşu). I choose as subject for my undergraduate thesis the sturgeon group, which allowed me to go the Danube Delta areas for few times.

In the evening, after a day of work on the beach, we gathered all on the famous boat placed on behind the station, together with Professor Sergiu Cărăuşu, who told us many stories about his travels on the seas from around the world, stories about fishes and important guests who came here every year from all academic centers of our country and abroad.

During morning, at sunrise, we went to fishery situated in front to see fishermen who bring different kind of fish in their bags. Sometimes, we were accompanying them in their boats. Leaving modesty aside for the moment, I remember Professor Sergiu Cărăuşu's appreciation, who told me that I have the same good knowledge about fishes as those who taught me.

I have already mentioned something about The Station Museum, which had an impressive number of exhibits preserved in formalin and many sheets with alga (all that could be found in the Black Sea). All students, after a rigorous training program, could present those exhibits to numerous visitors with relevant explanations. It was a huge satisfaction for us, especially for me, to be guides in the museum, to enhance the acquired knowledge. In that situation, we felt already as real naturalists.

Together with Professors Sergiu Cărăuşu and Mihai Constantineanu, we realized different trips in the surroundings of Agigea Station, especially around Agigea Lake. We learned there about its flora and fauna completely different than in the Black Sea. I remember a one-day trip made from Mangalia to Mamaia (both on beach and on waterfront) to know plants with flowers that grow on the coast, their adaptations to life on sand (psamophilic) or salty ground (halophyte) – a very abundant and interesting flora, from which, under the influence of anthropogenic factor, nothing rezisted! It was the year when it started the building of new hotels on the coast. The Park Hotel from Mamaia was the first.

During Sundays, we presented the life in the Black Sea. Our audience was formed by organized groups of pupils from scholar-camps. To prepare these reports, the resort library provided us treaties, determinators, books and many scientific journals. Every Sunday, Professor Sergiu Cărăuşu presented one episode about the life and work of the great Professor Ioan Borcea, founder of Agigea Resort, professor at University "Alexandru Ioan Cuza" from Iasi and member of Romanian Academy, founder of the Romanian school of oceanography.

Our evening walks were taking place on cliff and coast, from Agigea to Eforie Nord and back, following the pathway along the border of scientific reserve up to fishery and back. There was a total silence, hard to imagine nowadays with no radio, no TV, no music or clubs around. We were like ants with particular and clear missions, who came there to learn but also to relax, to become naturalists with good skills and competences, grateful followers of our professors who have formed us.

In this oasis, for which University of Iasi was enviable, is situated the famous scientific reserve with maritime dunes, founded by Professor Ioan Borcea. It has an impressive number of rare plant species, including *Ephedra distachya* and *Convolvulus persicus*. About the interesting flora placed in this natural reserve, I heard from our professor Constantin Burduja, who published several studies on coastal flora together with his collaborators: Adrian Volcinschi, Petrache Pascal and, especially, Clement Horeanu (who made a small botanical garden inside of Agigea

Station, which included the main species growing in Dobrogea District). Unfortunately, after 1970 when the Resort was passed under another administration, the garden was destroyed.

Concluding with these bits of memories about training stages at Agigea Station, I emphasize that I repeated these observations about flora, fauna and hydrobiology, during the summer of 1957. Since I graduated the university in 1958, until 1970, as preparator, assistant and lecturer, I annually returned in this area, leading training stages of naturalist students from the first academic year. In 1966 I participated to a national scientific session with international participation. On this special occasion, we celebrated four decades since this prestigious institute of science and education was founded, a part of "Alexandru Ioan Cuza" University, a famous institute with doors open to all institutions from our country, even for students or professionals from abroad. I returned to the station with great pleasure and interest as a botanist and ecologist. I was studying different coastal plant species, from morphological and anatomical point of view, including plants from scientific reserve. I entrusted to my undergraduate, postgraduate and doctoral students many scientific subjects concerning the arenarial flora of these habitats. Until 1970 I met here at Agigea new directors like Paul Borcea and Ionel Andriescu, famous professors at University of Iaşi, and new research-groups leaded by academicians Necrasov Olga and Petre Jitariu, Elena Jeanrenaud and Zicman Feider. They here realized a great research activity, widely appreciated.

After 1970, The Agigea station was a part of Romanian Institute of Marine Research. I will not refer to period 1970-1990 because I know it less. I rarely passed through the resort, but I registered gradually degradation of building, partial destruction of flora and fauna, change of pedo-climatic conditions, general appearance transformation when Agigea Seaport was built, reduced area where the station functioned, alarming "removing" of the Station from Black Sea, neglect and then cessation of fundamental research with practical implications. I noticed all these aspects related to this famous biological research institution, known and appreciated before 1970 throughout Europe and beyond!

*

* *

Immediately after 1989, the station from Agigea returned to the University "Alexandru Ioan Cuza" from Iasi, which founded it in 1926. The Faculty of Biology Council designated Hydrobiology professor Gheorghe Mustaţă as director. He is the person who retrieved and rebuilt it. In this period, here started again the scientific research and training stages activities made both by biologists and students from science and other faculties in the country. At the same time, were rebounded some old links with specialists from abroad, were recovered some buildings and utilities from IRCM Constanţa, have been made a number of new investments. In addition, the University has made some investments for comfort of its personnel who wanted and wants to spend holidays at the seaside.

Since 1991 at Agigea Resort it is organized every five years The Scientific Session of Faculty of Biology, with national participation, in collaboration with "Ovidius" University and Romanian Marine Research Institute from Constanta. In 1996 and 2001, as dean of faculty, I participated at the organization of these

scientific meetings. On these occasions, I have noticed that resort was returned to its status that it had until 1970, the improvement of conditions for student training stages (meals and accommodation), rearranging of laboratories, museum and Black Sea model, restoring of all green areas. Particularly, I have noticed the restoring of scientific reserve, expansion of rare plants which were in imminent danger of extinction.

Student training stages are run every year at the station, with access for students from other universities in the country and abroad. During summer time, professors, students, post-graduates students, PhD-students and researchers realize studies or collect biological material which is processed in laboratories of Faculty of Biology.

Since the beginning of this year, the management was entrusted to PhD Mircea Nicoara, professor of Hydrobiology at the Faculty of Biology from Iasi, who has a doctorate in marine biology. Personally, I appreciate the abilities of my students and colleagues to reorganize The Agigea Resort, their efforts to make it widely known and recognized, their interest in scientific research of Black Sea and Dobrogea District.

Today, at the 85th celebration of its existence, Station of Biological Research "Professor Ioan Borcea" from Agigea is among the great achievements of the University of Iasi. The Faculty of Biology could be proud of it, both as basis for education and research. I am talking about these four prestigious institutions: Natural History Museum (since 1834), Botanical Garden (since 1856), Biological Research Station from Black Sea (Agigea-Constanța, since 1926) and Biological Research Station from Eastern Carpathians (Potoci –Neamt, since 1956).

I have many wonderful memories about Agigea station, related to the students and their summer training stages, related to my graduate degree, all series of students which were here together, related to my first phyto-anatomical researches which I made. But above all, it remained imprinted in both my memory and my heart, this academic ambiance for study founded there. Also, I have in memory the biology major Romanian personalities that I met there as a student and, later, as professor of illustrious *Alma Mater Iassensis*. I still think about the ones I have heard here in Agigea: Țuculescu, Celan, Şanța, Codreanu, Pora, Jitariu, Necrasov, Papp, Burduja, Jeanrenaud, Constantineanu, Feider and, not the last, the famous professor and director Sergiu Cărăuşu.

During the six decades since I have worked at the University of Iasi, I had the opportunity to know many biological research units from Europe and elsewhere, but none of them was so close to my soul that Agigea Station. Here, more than anywhere else, we have learned to know and to love nature. We met here many great biologists, models for me and my colleagues during those years that followed. I am confident and optimistic that the coming days will allow to be rebuilt here a permanent staff of researchers. The new director, Professor Mircea Nicoară will be able to combine his enthusiasm and experience with scientific spirit of the ancestors. Agigea Resort was, is, and must remain a sanctuary of Romanian biology, an oasis full of tranquility and charm, a vibrant seaside atmosphere of today.

"PROF. DR. IOAN BORCEA" MARINE BIOLOGICAL SCIENTIFIC STATION FROM AGIGEA

GHEORGHE MUSTATA
"Alexandru Ioan Cuza" University of Iasi, Faculty of Biology, Bd. Carol I, 20A, 700505, Iasi, Romania

There are 85 years since the establishment of the "King Ferdinand I" Maritime Zoological Station from Agigea. By the Royal High Decree no. 810/1st of March 1926, it was decided both the establishment of station and appointment of the university Professor Ioan Borcea, as director.

Professor Ioan Borcea's appointment was not a random decision. The station establishment was mainly due to the efforts made by professors of his scientific and organizational authority and of high-ranking relationships at the Royal Court and at the Ministry of Education. Honouring, in 1919, the function of Minister of Education, he saw his dream come true: the building of foundations of a marine research station on the Romanian Black Sea coast. Performing hydrobiological research in the Black Sea and in its lagoons, he realized how important can be such a scientific station.

To have success in his efforts, he carried out a scientific expedition from the Danube mouthes to Vama Veche in 1925, the main objective being the finding of a good location for this station. The Professor aimed at two main aspects: the ensuring of a basis of material reception and a place in which the marine lanscape should be as much varied as possible. Such is explained the fact that he chose as place for the foundation of the scientific station a coastal zone near the locality Agigea. Here were found military barracks which functioned in The First World War, with buildings that met the proposed goal, and coastal marine ecosystems were very different, offering the possibility to study hydrobionts in all types of facieses.

Accumulating all the necessary data, the professor realized an organization and functioning project for the first Romanian Scientific Station of Marine Biology. He accumulated required experience in France at the Sorbonne, and at Banyuls-sur-Mer and Roskoff Stations. The great advantage for the position of the Station was the fact that it was located between the Black Sea, Techirghiol Lake and Agigea Lake.

Being endowed with an unusual power of work and bestowed by God with a talent of a great diplomat, Professor Ioan Borcea succeeded to persuade the higher forms of necessity for such a station for biological research on Romanian Black Sea coast and demonstrated that the place is the best choice for such an institution. This does not mean that he did not have emotions in expectation of coming out of the Royal Decree. Also then, in same maner as now, there were many officials to delay things.

However, on March 1st, 1926, the Higher Royal Decree appeared and professor saw his dream fulfilled – the establishment of Station and the utilization of material basis of barracks.

Professor's qualities of founder proved efficiency beyond measure. A surface of about 25 ha, located 10 km away from Constanta and near Eforie Nord,

was surrounded by a fence, of which some parts are also functional today, marking Research Station territory, and thus became residence of the "embassy" of University of Iasi in Constanta. Barracks buildings were a starting point and were used as appropriate for research in Marine Biology and biological practice of students.

For the beginning, six fishermen and an administrator were employed and a means of transportation was bought for movement on sea and processing of scientific material. The first scientific expedition was organized on Black Sea in 1926. The second expedition was organized in 1928, with "Posidonia" ship, reaching to Kaliakra Cape. Whole scientific material collected passed through hands of the professor, was prepared and processed scientifically, stopping himself more on some groups, going up to identification of main species. His taxonomic researches were oriented to fish and namely to families: *Gobiidae, Clupeidae, Bleniidae* and *Labridae*. He also did researches on crustaceans and molluscs. At the beginning, being alone, he tried and managed to know all groups of hydrobionts, and then he began to initiate some of his students, who later became his collaborators. It is about Academy member Mihai Băcescu and Professor Sergiu Cărăuşu.

Native intelligence and his great power of work were paradoxically joined with a strong personality, often feared and with social valences worthy of admiration, that is why he was loved by students and collaborators. He knew so well Black Sea fauna, so that he impressed all those who were coming to Agigea by his knowledge. He was finding answer to all problems of Black Sea. Recognition of common species from all systematic groups, their association according to benthic facies, geographical distribution of some species and even some problems concerning the origin of Black Sea fauna. He was inviting some of his better students to help him in sorting scientific material and to initiate them in problems of Marine Biology.

In a short time, Scientific Station from Agigea has become a major attraction for great Romanian biologists, but also for those who want to know mysteries of Black Sea. Having enough accommodation space, it began to polarize around it the greatest zoologists, botanists, geologists, chemists and physicists, and scientific discussions held on threshold of evening were becoming real true lessons of Biology. Many of the greatest zoologists were specifically taught by Professor in order to broaden spectrum of discussions. In fact, in 1928, The First Congress of Naturalists invited not only zoologists, but also botanists to visit Agigea, stirring up their interest for rare and endemic plants in Dobrogea and even for those inside of station. The greatest botanists of the time responded to his invitation: Alexandru Borza, Iuliu Prodan and Erasmus Nyarady. They studied the plants on Agigea dunes and discovered a new species, which was dedicated by E. Nyarady to Alexandru Borza, who discovered it – *Alyssum borzeanum*, which now forms the whole meadows.

Confirming itself, plant richness characteristic to marine dunes, Professor Ioan Borcea enclosed a surface of 600 m^2, where density and variety of plants were higher and named it Protected Area of Marine Dunes from Agigea. Professor's dream became a reality thanks to efforts made by Alexandru Borza, who was at that

time President of Nature Protection Committee, and who officially declared in January 1939 a surface of 6300 m^2 as Protected Area of Marine Dunes.

Meetings from Agigea could be compared with those from Iassy within Junimea Literary Talks. Having planted in his soul "sacred fire" of scientific research, Professor Ioan Borcea made from Agigea Scientific Station the largest Romanian school of marine biology. Much has been said and is still spoken of „Spirit from Agigea" enthroned by Professor Ioan Borcea.

Professor Ioan Borcea proposed himself to make from Agigea Scientific Station a school for the Romanian biologists. He was named "Titan of Romanian Zoology". Professor considered that you can not achieve anything in field of scientific research if you do not wear in you the "sacred fire" and do not work with passion and dedication, without thinking of immediate profit or material gain.

Professor had cult of work and spread around him thirst for knowledge. He was attracting students and specialists around him like a lighthouse in the night. In fact, Academician Petre Jitariu wrote in 1979 on the occasion of centennial of professor's birth: "Therefore, Agigea represented attraction point for those who had in soul and conscience those torches that had to be lightened at fire of unquenchable desire to grow according to loved and admired Professor's model by those who knew him and worked under master's leadership, full of wisdom and warmth".

This Scientific Station has become a real Mecca of the Romanian biologists. This did not represent just a Station at the Pontus Euxinus Seaside, but a School of Marine Biology and more than that, a state of spirit.

Based on his encyclopaedic spirit, Professor Ioan Borcea has developed several research directions in the field of marine biology:

- knowledge of Ponto-Caspian relics in banks of Dobrogea and Black Sea;
- elucidation of migratory fish origin from Black Sea;
- taxonomic researches on *Gobiids, Mugilids,Clupeids* and *Bleniids*;
- researches of morphology and comparative anatomy on *Gobiids*;
- elucidation of Black Sea fauna origins;
- the study of crustaceans and molluscs;
- researches on the structure of benthic biocoenosis in comparison with nature of populated facieses;
- elucidation of some aspects concerning the fauna of estuaries in Dobrogea in connection with adaptations determined by transition from brackish environment of Black Sea to freshwater one.

Professor Borcea fell like a thunderbolt in 1936, on July 30, as a result of a septicaemia caused by a working table in laboratory. Professor's loss was covered by his scientific papers, by Schools of Biology he founded: Marine Biology, Animal Morphology and Entomology.

The most precious treasure which the Titan of the Romanian Zoology left us is his soul foundation, The "King Ferdinand I" Maritime Zoological Station which has survived in the course of time and of times, reaching up to 85 years, the moment of our anniversary.

Critical moment, in which this Scientific Station was found, has been overcome by the appointment of Professor Constantin Motaş, as a Director. A real

grey eminence, Professor Motaş was a specialist of European renown in Hydrobiology domain and one of the greatest hydroecologists of the world. By special efforts, he has succeeded to equip Agigea Station with "Sagitta" motorboat, to equip two zoological research laboratories and to maintain at a high rate "Agigea's Spirit". Agigea Scientific Station has remained just as welcoming as in Professor Borcea's time. A significant achievement is the establishment of a scientific journal of the Station called Works of "King Ferdinand I", Marine Zoological Station, and to coordinate printing of the first 3 numbers (1938, 1939, 1941).

After departure of Professor Constantin Motaş to Bucharest, in 1941, it was appointed as director, Professor Ioan Gh. Botez, one of the greatest anthropologists of all times. The greatest merit of new director was that he has succeeded to maintain Agigea Station working in that black period during Second World War. Along with Director, there were also found Mr. Cărăuşu Sergiu and Mrs. Cărăuşu Aurelia, who were employed as researchers.

In 1949, after passing of war wrath and radical transformation in Romanian politics, delators of Professor Ioan Gh. Botez managed to give him on Security hand, he was arrested and taken to Danube-Black Sea Canal from where he did not return any longer, dying as a martyr. At management of Agigea Station followed Lecturer Sergiu Cărăuşu who later became Reader and Professor.

The reform of education, which took place in 1948, determined changes of the Station name, this being dedicated to the memory of the founder - "Professor Ion Borcea" Marine Zoological Station.

Evolution of Scientific Station has had an upward direction; number of researchers has grown considerably. Florica and Ioan Porumb couple and Octavian Şerbănescu to whom joined Maria Celan - one of the greatest Romanian algologists - and others considerably raised the scientific prestige of the scientific station by research laboratories established and led by them, through value of published works.

Little by little and step by step, Agigea Research Station has become a true Institute of Marine Biology. In the period 1962-1966, there were employed 12 researchers, considerably widening research spectrum approached: - zooplancton – PhD. Florica Porumb; - Ichthiology – PhD. Ion Porumb; - marine phytoplancton – Pia Mihnea; – marine zoobentos – Ionel Andriescu, Radu Mihnea, Dan Manoleli; – Vegetal Physiology– Maria Stadniciuc, Aurora Ciobanu; - Animal Physiology – Valer Crăciun, A. Telembici, Ioan Neacşu; - Radiobiology – Octavian Şerbănescu; – Terrestrial Ecology – Victor Ciochia, Carol Nagy, Clement Horeanu.

The large Agigea Station building was completed with a row of rooms in the part to Black Sea; it was so successfully built the model of Black Sea. Agigea Scientific Station had two research ships that effectuated long expeditions intended for knowledge of coastal ecosystems. „Emil Racoviță" ship was transferred to „Stejarul" Biological, Geographical and Geological Station from Pângărați.

At 30 years after its establishment, Station had 12 laboratories: 5 laboratories of Zoology, Anatomy and Embriology, Biological Chemistry, Botany, Microbiology, Animal Physiology, Chemistry and one student's laboratory. Laboratories were equipped with modern equipment.

Agigea Station Library included over 7000 books and received 240 journals of speciality, by exchange. At leadership of Station the following persons succeeded: Professor Paul Borcea, Lecturer Ion Suciu, Professor Nicolaie Gavrilescu and Professor Ionel Andriescu.

In 1970, the Agigea Scientific Station was in full glory. It became a Marine Research Institute, had already passed under direct coordination of Ministry of Education.

Spectrum of researches was very varied and professors and researchers across country were coming to Agigea Scientific Station.

School of Animal Physiology of Academy member Eugen Pora worked along side with that of Academy member Petre Jitaru from Iaşi. Concerning the researches of physical and chemical characteristics of Black Sea waters, a special contribution had: C. V. Gheorghiu, N. Calinicenco, Radu Cernătescu, A. Mangeron and others.

In 1969, between July 15 and 31, organization of MAMBO course took place under the aegis of The Mediterranean Association of Marine Biology and Oceanology, with support of FAO, at which many persons attended: 22 researchers and professors from 11 countries and 21 professors, readers and Romanian researchers, among which Academy member Eugen Pora, Academy member Mihai Băcescu, Professor Constantin Motaş, Professor Radu Codreanu, Reader Maria Celan, etc.

During the glory period of Agigea Station, when it was affirmed on scientific plane and it was been included on big similar European Scientific Stations List, disaster was produced.

Although "Prof. Ion Borcea" Marine Biological Scientific Station and "Grigore Antipa" Institute of Oceanography were functioning in parallel, without overlapping in scientific subjects, and were functioning well, paltry interests and desire of upstarts of some researchers led to merging of two research institutions and formation of Romanian Institute of Marine Research. These two institutions have been not just under one leadership, following to continue their researches and to increase their efficiency through collaboration, but fusion was done by reducing number of researchers, thus the team from Agigea being disorganized. By 1975, research was still done at Agigea. Alongside of those remained under new organization, researchers from Constanța came too, some of them have become great personalities of Romanian Biology and after withdrawal of Romanian Institute of Marine Research from Agigea and concentration of all researchers in Constanța, Agigea Station just practically disappeared from Romanian research.

The recovery of Station

For nearly 20 years, Agigea Station has been invaded by a number of institutions related to maritime and port activities: SERUN, which belonged to ICHP Constanța, Constanța Shipyard, CUG Basarabia and a Military Construction Unit. Like Pepelea's nail in one of former Station buildings, Reader PhD. Maria Celan, the greatest Romanian algologist has remained to continue her existence. By her personality and behaviour, Maria Celan has still succeeded from time to time to wake up leaders of invading institutions to reality and to draw their attention on

importance and significance of Protected Area of Dunes from former Station perimeter. Like a ghost, Maria Celan has filled space-time between two great events: abolition and Station recovery. Leaving us in summer of 1989, she has not been able any longer to see her Station saved. We have presented in previous reports events that have led to recover the Station by „Alexandru Ioan Cuza" University.

We express our duty to remember the necessary steps taken by Dean of that time, Professor PhD. Constantin Pisică, and especially of "Alexandru Ioan Cuza" University Rector, Professor PhD. Călin Ignat. The recovery could not been achieved without support of two great personalities: Professor PhD. Adrian Rădulescu, Rector at that time of "Ovidiu" University and Mihai Dulică, General Director of ICHP Constanța.

Rector Călin Ignat was friend with Rector Adrian Rădulescu, who was at the same time Prefect of Constanta, too. Added to this, after a few visits paid to Director Mihai Dulică (may God reward his deeds in His Kingdom!), a sincere and disinterested friendship was realized between us. We can affirm that Director Mihai Dulică took on his own account the transfer and personally took care of getting it.

We have to bring our thanks both to Professor Adrian Bavaru, Dean of the Faculty of Biology at that time, then Rector (in two legislations) and to Director of the IRCM Mr. Simeon Nicolaev, who agreed with recovery of Agigea Station.

On the 28th of June 1990, "Prof. Ioan Borcea" Marine Biological Station has officially returned to "Alexandru Ioan Cuza" University based on transfer order no. 4147/28 June 1990, achieved between Ministry of Transport and Telecommunications and Ministry of Education.

„Prof. PhD. Ioan Borcea" Marine Biological Station has been recovered. What namely was it recovered? There were recovered buildings and a part of territory of the former Scientific Station.

It was not possible to recover anything in what concerns library and its patrimony, not even Lucia Leon Borcea's donation (a library with historical and sentimental value).

Though in summer of 1990, I did the first practice with students at Agigea Scientific Station, receiving a substantial support from Director Mihai Dulică in arrangement of accommodation space, The SERUN's late withdrawal (in December) without heating buildings with inundation caused by freezing of some pipes produced a serious deterioration of large building by inundation. However, until 29 of June 1991, when it was make official opening of Agigea Scientific Station, there were made repairs to three buildings and it was arranged accommodation space for over than 100 students and about 20 teachers and researchers.

It was ensured equipment and continuous operation during summer. Practically, Agigea Scientific Station has become functional again. All efforts made have been oriented towards providing optimal conditions for summer students practice.

There have been resumed the first scientific researches in Black Sea coastal ecosystems. There were employed the first researchers: Mariana Parincu and then Aurelia Balaban.

Organization of Agigea Scientific Station approved in 1990 at putting into function of Agigea Scientific Station, comprising: - one administrator- 4 guards- 4

stokers- 5 researchers- 2 skilled workers– 2 unskilled workers (for maintenance Protected Area with Dunes inside of Agigea Scientific Station).

The most complete scheme was in 1995:

- Director – Professor PhD. - Gheorghe Mustață;
- Administrator - Osman Ural;
- Researchers - Pălici Cristina, Costin Junona, Surugiu Victor;
- Duty women - Dobre Ioana, Turlacu Zenovia;
- Guards - Bozian Corneliu, Stoican Florin, Videanu Valentin, Nechita Constantin ;
- Firemen - Arif Ermin, Talabă Gheorghiță, Chițcanu Ion;
- Electrician - Drăghici Ioan;
- Plumber - Decu Costică ;
- Unskilled workers - Maria Bucur, Nistor Vasilica.

There was a fairly large circulation of researchers at Agigea Scientific Station. Among researchers who worked in Agigea Station, we mention:

- Parincu Mariana – 1991-1994;
- Balaban Aurelia – 1992-1995;
- Trandafirescu Iulian – 1993-1999;
- Costin Junona – 1995-1998;
- Pălici Cristina – 1995-2007;
- Surugiu Victor – 1995-1996;
- Staicu Ivona – 1998 – 1999;
- Cojocariu Cristian – 1998 – 1999;
- Ursu Felicia Olga -1998 – 1999;
- Micu Sânziana – 2005 – present.

In 1999, 5 researchers were working in Agigea Scientific Station, but, in the same year we were forced to reduce their number by 3 because Ministry of Education and Research did no longer accept employment of researchers in universities.

Number of researchers has gradually decreased because it was not expected a perspective of a research career at Agigea Scientific Station. Three researchers went since 1999, and then another one, only an assistant researcher remaining: Cristina Pălici, who, after she has successfully passed her PhD. stage, went to England. After her departure, the Station had no researchers. The situation could be somewhat saved by orientation of two my doctoral students to marine research. To facilitate their activity, I have offered them accommodation inside of Agigea Scientific Station space and laboratories for research. It is about Sânziana Micu and Dragoş Micu. Mrs. Sânziana Micu (married Ghițan) completed doctoral thesis and was then permanently assigned as a researcher at Agigea Scientific Station.

There is one researcher only at Agigea Scientific Station (Dragoş Micu transferred to IRCM), but being now on vacation for birth, we have to consider, in fact, that there is no scientist.

We use this opportunity to request to the management of "Alexandru Ioan Cuza" University at least 3-5 researchers to form a working group able to operate a contractual activity to bring necessary funds for research – development. By taking

over Agigea Scientific Station, we have moral duty of renewing broken threads of research from Agigea.

I had to resume research activity from zero. First researchers have been taken on staff and took action. At first, because of lack of means of navigation on Black Sea, we had to orient our attention on superlittoral and mediolittoral. The first researches stipulated by contract have started. Then, equipped with "Noctiluca" and "Rissoa" motor boats, we began to detach from shore.

In 1995, a Romanian-British expedition was organized in Razelm Sinoe lagoon complex and in the Black Sea. Application was organized together with experts from Oceanography Department - University of Southampton, namely: Paul Riddy-geophysicist; John Williams–planctonologist and Martin Sheader–benthologist. From the Romanian part participated in this expedition: Professor Gheorghe Mustață, researcher Iulian Trandafirescu, Assistant-researcher Victor Surugiu, Assistant-researcher Cristina Pălici and Professor Assistant Mircea Nicoară.

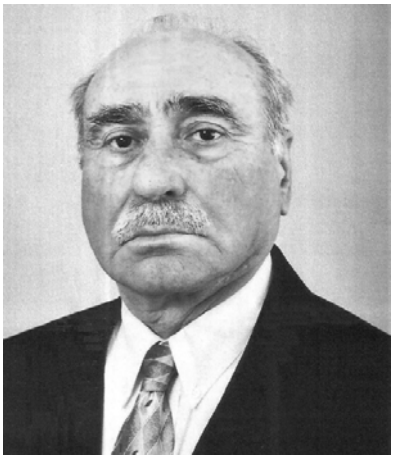
We believe that this expedition has constituted a moment of truth. Specialists from Agigea Scientific Station were able to organize such an expedition and to complete researches by several published papers. The success of the expedition was appreciated by specialists from Southampton, and in response, for 5 years, students and researchers from “Alexandru Ioan Cuza” University have received study scholarships and worked in the laboratories of Oceanology Department from Southampton University.

We have started with a small group, however, to continue some of researches effectuated at Agigea. Since 1991, we have realized the first research contract "The influence of pollution and the Danube-Black Sea Canal on Black Sea hydrobionts".
A series of contracts followed by which we have succeeded to re-enter in circuit of research such as:

Programme/ Project	**Function**	**Period**
Program of integrated actions (PAI) "Brâncuşi" signed on 26 March 2002 between the MEC and French Ministry of Foreign Affairs – for French part. Project NEANTHES – Approche multidisciplinaire pour l'évaluation des impacts de la pollution chez deux espèces clés de la Mer Noire	Responsible – Romanian part	2007-2008
Project of arrangement within the Funding Program for the Cycle of licence Laboratory of Ecology, Geography, Physics, Chemistry and the Biology of the Black Sea - 87. 170 RON	Director	2007
Contract 193-Consortium with INCDM and the "Babes-Bolyai" University from Cluj-Napoca-Biological and genetic biodiversity of the populations of *Rapana venosa* from the Romanian Black Sea coast - 270. 000 RON	Coordinator from "Alexandru Ioan Cuza" University	2006-2008

THE LOSS OF A GREAT ICHTHYOLOGIST OF OUR NATION – PRINCIPAL RESEARCHER I DR. IOAN PORUMB (1925-2011)

GHEORGHE MUSTATA
"Alexandru Ioan Cuza" University of Iasi, Faculty of Biology, Bd. Carol I, 20A, 700505, Iasi, Romania



The anniversary moment devoted to celebration of 85 years since the establishment of „Professor Dr. Ioan Borcea" Marine Biological Station is marked by a painful event, namely passing of man who was one of the great Romanian ichthyologists who did research at Black Sea, principal scientific researcher I PhD. Ioan Porumb.

A traveller in two centuries and two millennia, principal researcher I PhD. Ioan Porumb is for us, together with his wife, Mrs. principal researcher I PhD. Florica Porumb, a symbol, top of the iceberg, through which the "Prof. PhD. Ioan Borcea" Marine Biological Station is immortally maintained in twenty-first century too. The two were hired at Agigea Station in 1949 and have dedicated their whole life to Marine Biology researches. Mr. and Mrs. Porumb became frontally employed in research, going in tandem, or separating them, according to approached research theme. Principal researcher I PhD. Ioan Porumb has dedicated his life to knowledge of Black Sea ichthyofauna and not only. Continuing the research of his great predecessor Professor PhD. Ioan Borcea, he has covered other aspects of research in Ichthyology, such as elucidation of nature of food for many species, knowledge of their biology and spread in Black Sea according to food sources and migration that some species carried out.

His researches were oriented too towards valuation of some fish stocks with commercial importance. Such researches have had a major economic importance for development of fishing economy in Black Sea area.

Achieving excellence marine biology research, principal researcher I PhD. Ioan Porumb, had distinguished himself, as I affirmed, as one of the greatest ichthyologists of our nation. You cannot do ichthyological research at Black Sea without knowing the published papers by the illustrious scientist, professor Ioan Borcea. The fact that, together with his wife, Mrs. PhD, Florica Porumb, have effectuated research concerning the structure and seasonal dynamics of nyctemeral zooplankton allowed him to elucidate many aspects regarding the biology and migration of planctonophagous fish.

Principal researcher I PhD. Ioan Porumb was born in Iasi on the 9th of November 1925. Childhood and school, up to academic level, were also related to

Iasi. He attended classes of primary school at "Cuza Vodă" School, inferior course at „Costache Negruzzi" Boarding High School and the superior course at National High-School. He successfully graduated baccalaureate in 1944, but country being at war, he had to go as soldier on front and at the end he was promoted to rank of officer.

After war, he attended courses of the Faculty of Sciences, Department of Natural Sciences, which he graduated in 1948.

The young graduate was appointed as a Biology teacher at Gymnasium in the Ciurea District. But, distinguishing himself among his colleagues by native intelligence, a great power of work and a very particular steadfastness, he was called as a university Professor Assistant at university and then was transferred as a scientific researcher at "Prof. Ioan Borcea" Marine Biological Station on the 2nd of February 1949.

At that time, Prof. PhD. Ioan Gh. Botez was Director of Agigea Research Station. He worked here until 1970, when the Station was abolished, occupying the functions of principal biologist, principal scientific researcher and the Head of Ichthyology Department.

Principal Researcher I PhD. Ioan Porumb carried out ichthyological researches with a broad spectrum coverage, researches of biodiversity:

- biology and ecology of some species;
- migration of some species;
- estimation of fish stocks with commercial importance.

Important contributions were brought as concerns biology of some pelagic genera: *Alosa, Atherina, Belone, Blenius, Engraulis, Gobius, Odontogadus, Pomatomus, Sarda, Sardina, Trachurus*, etc.

Trofophological interesting research was done on several species of fish. A novel method was developed for recognition of species that serve as food, depending on the somatic still undamaged fragments of digestive enzymes.

Following the list of scientific papers published in those over 60 years of work, you remain impressed by the manner in which the Mr. and Mrs. Porumb worked. One may think that zooplankton is a research direction that has little in common with ichthyology domain. Nothing falser, so much as many species of fish are planctonophagous.

Two of papers devoted to zooplankton study lead to make a synthesis paper regarding to the zooplankton–fish relationships:

- Porumb F., Porumb I., 1965 – Recherches concernant la migration nycthémérale du zooplancton marine d'été. –Rév. Roum. Biol., sér. Zool., Ed. Acad. R.S.R.; (Research regarding to nicthemeral migration of summer marine zooplankton);
- Porumb F., Porumb I., 1966 – Cercetări asupra migrației nictemerale a zooplanctonului marin de vară. St. Cercet. Biol., ser. Zool., Ed. Acad. R.S.R.; (Research concerning to nicthemeral migration of summer marine zooplankton);
- Porumb F., Porumb I., 1982 – Base trophique zooplanctonique et son importance pour la répartition et la nutrition du sprot dans les eaux devant la litoral roumain de la Mer Noire. (Trophic

zooplanktonic base and its importance for distribution and feeding of *Sprattus sprattus* species in the waters in front of Romanian Seaside). Cercetări Marine, I.R.C.M. Constanța.

In the support of this idea we mention the following paper: Porumb F., Porumb I., 1983 – „Importance du zooplancton pour la répartition des poissons pélagiques dans les eaux du plateau continental devant le littoral roumain de la Mer Noire. Rapp. Commm. Int. Mer Medit., C.I.E.S.M. „The importance of zooplankton for distribution of pelagic fish in waters of continental plateau in front of Romanian Seaside of Black Sea.

We especially appreciate researches regarding the ecology of some fish species. These studies investigated populations structure of *Sprattus sprattus, Engraulis encrassicholus ponticus*, etc.

The ability to realize scientific coverage and documentation of principal researcher I PhD. Ioan Porumb allowed him to do, along with some collaborators, synthesis papers of great value, with scientific and practical significance:

- Porumb I., Iordănescu V., 1984 – Ressources piscicoles des eaux du plateau continental roumain de la Mer Noire et la perspective de leur mise en valor. Cercetări marine I.R.C.M. (Piscicultural stocks of waters of the Romanian continental plateau of the Black Sea and the perspective to put them into value);
- Porumb I., Iordănescu V., Papadopol N., 1980 – Aspecte privind perspectivele economiei pescăreşti la Marea Neagră. Viitorul mărilor şi oceanelor (M. Malița, Ed. Acad. R.S.R.) (Aspects regarding the perspective of fishing economy from Black Sea).

The biodiversity of fish is still in decline in the Black Sea. Comparing the current situation with that existing in 1981, one can truly understand what is the reality of our days (2012). It is necessary, however, to follow the research of the principal researcher I PhD. Mr. Ioan Porumb whose results were published in several papers. We here nominate only two:

- Porumb I., Marinescu F., Butoi G., 1991 – Variations of the composition of the pelagic fauna in the Romanian fishing of the Black Sea. Comm. Int. Mer. Medit., C.I.E.S.M. Monaco;
- Porumb I., Marinescu F., Gorban A., Butoi G., 1979 – Data regarding the evaluation of stocks of some species with economic importance in the waters above the Romanian continental platform at the Black Sea. Ses. şt. jubiliară. Fac. de Tehn. şi Chimia prod. alimentare şi tehnica pescuitului, Galați.

Principal researcher I PhD. Ion Porumb was among researchers who passed from Agigea Station to Romanian Marine Research Institute, in 1970. Here, he had the function of Head of laboratory. This passing did not affect with anything excellence of researches. However, something happened: He drew around him other researchers too, he guided their steps in Ichthyology, and some papers were made in collaboration.

We want to mention that during 1950-1959, researcher Ioan Porumb also acted as Professor Assistant at Ichthyology, Physiology and Anatomy Department at

Institute of Fisheries and Pisciculture, and between 1954-1955 was scientific researcher at Commission of Oceanology of Romanian Academy.

After retirement, in 1988, he has continued his activity as a consultant researcher.

He carried out an intense activity also in his quality as member of Navy League of Romanian Constanta branch, where he co-ordinated The Department of Environment, and as a member of editorial staff of The Journal Our Sea.

At the beginning, I affirmed that Mr. and Mrs. Porumb formed a brilliant tandem both in research and in everyday life. Many of last papers are signed together – Porumb Ioan & Porumb Florica.

It is a pleasure for me to present here THE MAN Ioan Porumb - as wonderful and special as he has been. But I have not known him enough. However, I felt in 1991-1992 how devoted to his family he is. He spoke to me with pleasure and with justifable pride about his son working within Romanian Post.

Principal researcher I PhD. Ioan Porumb physically left us, but he did not leave us totally. He left behind a valuable scientific work and a certain model of life. At this time, we are alongside with Mrs. Principal Researcher I PhD Florica Porumb and his son and they would like, in order to keep him in our memory, to offer us some of his personal objects which are to be exhibited in Agigea Station Museum.

Mrs. Florica Porumb has remained Dean of age of researchers who have worked at “Prof. Ioan Borcea” Marine Biological Research Station. We wish her good health to commemorate her husband and also to fully feel warmth in our soul.

TIPARUL EXECUTAT LA
IMPRIMERIA EDITURII UNIVERSITĂȚII
„ALEXANDRU IOAN CUZA“ DIN IAȘI
700109 Iaşi, Pinului 1A, tel./fax 0232 314947
Apărut: 2013
Comanda:186

Informații și comenzi:
www.editura.uaic.ro
editura@uaic.ro