

## WATER QUALITY ASSESSMENT IN THE RAŠKA RIVER BASED ON ZOOBENTHOS AND ZOOPLANKTON ORGANISMS AS BIOINDICATORS

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### KVALITET VODE U RECI RAŠKOJ NA OSNOVU ORGANIZAMA ZOOBENTOSA I ZOOPLANKTONA KAO BIOINDIKATORA

#### *Apstrakt*

U cilju ispitivanja kvaliteta vode reke Raške, organizmi zoobentosa i zooplanktona korišćeni su kao bioindikatori. Istraživanje je sprovedeno od aprila 2011. godine do maja 2012. godine, u vremenskim intervalima od dva meseca. Odabrano je ukupno pet lokaliteta na dužinini toka od 2.5 km. Između drugog i trećeg lokaliteta lociran je pastrmski ribnjak, čiji je uticaj na zajednice organizama praćen. Na lokalitetima koji su obuhvaćeni istraživanjima konstatovano je 57 taksona makrozoobentosa (34 vrste, 21 roda, 1 familija i 1 klasa) i 75 taksona zooplanktona (58 vrsta, 15 rodova, 1 red i 1 klasa). Srednje vrednosti indeksa saprobnosti ukazivale su na manje razlike u dobijenim vrednostima korišćenjem zoobentosa i zooplanktona. Na osnovu saprobioloških analiza, kada su kao bioindikatori korišćeni organizmi bentosa, kvalitet vode u reci Raškoj je druge klase kvaliteta, ili na prelazu između prve i druge klase, uglavnom na lokalitetima iznad ribnjaka. Organizmi zooplanktona su pokazali da voda celom dužinom toka pripada prvoj klasi kvaliteta. Najmanje vrednosti indeksa saprobnosti zabeležene su na prvom, referentnom lokalitetu  $1.453 \pm 0.098$  (zooplankton), a najviše na četvrtom lokalitetu  $1.88 \pm 0.021$  (zoobentos). Na lokalitetu ispod ribnjaka, gde je zabeležen pad u koncentraciji kiseonika, dominiraju organizmi bentosa koji tolerišu veći stepen organskog zagađenja (Chironomidae, Oligochaeta, Simuliidae). Iako su koncentracije ukupnog fosfora i ortofosfata rasle od trećeg ka petom lokalitetu, njihove koncentracije nisu premašivale dozvoljene vrednosti za salmonidne vode. Međutim, srednje vrednosti koncentracije nejonizovanog amonijaka ( $\text{NH}_4^+$ ) su iznad referentnih vrednosti na trećem i četvrtom lokalitetu.

Razlog nepodudaranja vrednosti indekasa saprobnosti za dve istraživane grupe, može biti usled nestabilnih zooplanktonskih zajednica zbog izraženog efekta drifta, kao i usled činjenice da će zbog procesa sedimentacije dospelih organskih materija, organizmi dna biti izloženiji njihovom uticaju. Zajednice bentosa, kao znatno stabilnije i u vremenu i u prostoru, pogodnije su za procenu kvaliteta tekućih voda.

*Ključne reči: zoobentos, zooplankton, kvalitet vode, bioindicators*

*Keywords: zoobenthos, zooplankton, water quality, bioindicators*

## INTRODUCTION

Freshwater pollution represents one of the major global problem of the modern age. Various human activities, particularly those related to agriculture, have a great impact on the amount of organic matter found in freshwaters. In Serbia, some first and second order streams are under the influence of trout farms that are built close to their sources. Organic matter in the form of degraded fish feed and feces are drained into the environment usually without previous treatment and have a big impact on water quality of the recipient (Marković and Poleksić, 2011). Chemical parameters, as indicators of water quality, do not provide the state of biological communities and therefore cannot properly indicate the status of freshwater ecosystems (Camargo *et al.*, 2011). Over the last decades, biological methods have been recommended as a useful tool for the assessment of water quality (Hynes, 1970). Together with chemical parameters, they are used in monitoring freshwaters.

In addition to various types of biotic indices, saprobiological analysis of water ecosystems is one of the main methods for the assessment of water quality using aquatic organisms. Each water quality class has a corresponding saprobic state based on the amount of certain organic material susceptible to degradation (Moog, 2002). Different groups of animal and plant species such as algae, macrophytes, zooplankton, macrozoobenthos, meiobenthos and fish can be used in biological monitoring.

In first and second order streams biological monitoring, is usually based on zoobenthos as the most stable and the most diverse of all aquatic communities. In many lotic systems they form communities with over a hundred species from many phyla such as Arthropoda, Mollusca, Annelida, Nematoda i Platyhelminthes (Turbellaria) (Hose *et al.*, 2004). As a group also used in biological monitoring, zooplankton offer several advantages: they have worldwide distribution and their communities are sensitive to changes in environmental factors as well as the amount of organic matter in aquatic habitats, therefore can indicate water quality (Arimoro & Oganah, 2010). However, zooplankton is not often used in monitoring especially in smaller rivers, due to intense drift effect on its communities. Though, many studies show that they may be present in such water bodies, particularly within the phylum Rotatoria. According to Kobayashi *et al.* (1998) flow velocity is the main factor affecting the diversity of zooplankton in lotic systems.

The aim of this study was to determine the effects of a trout farm on the water quality downstream from the recipient, using macrozoobenthos and zooplankton as bioindicators. Additionally, we compared the values of saprobic index and chemical parameters in order to determine the level of correspondence between the results.

## MATERIAL AND METHODS

### *Study area*

The Raška River is a left tributary of the Ibar River, located in the southwestern part of Serbia. Its length is 39 km, width 10-25 m, depth 0.3-1 m and the catchment area is 1040 km<sup>2</sup> (Gavrilović i Dukić, 2002). The source of the Raška River is a specific geomorphological phenomenon. Karst Koštam field is a shortened source of the river Raška and is located 950 m a.s.l. (Marković, 1988). Across the bottom of the Koštam field are sinks of the Točilovska, Delimeđska and Likovska River. Waters of Koštam field are drained and flow through underground channels to emerge from the cave in the form of a strong spring, in the vicinity of the monastery Sopoćani. This water is used for electricity production and partly for trout farm supply in the area of Ras, in the village Pazarište.

This research was conducted during 2011 (April, June, August, October, December) and 2012 (March and May). Sampling of macrozoobenthos and zooplankton was carried out at five localities of a channel formed by redirecting the Raška River from its source (2.5 km in length). (Figure 1).

The first locality (RŠ1) is situated near the river source at 557 m a.s.l. RŠ2 locality is 400 m away from the RŠ1, at 548 m a.s.l. RŠ3 locality is placed 10 m below the outlet of the channel owned by the trout farm "Vekoslav Vukićević". Communities of aquatic macrophytes are dominant at this site. Two hundred meters below the third locality is RŠ4 locality, on 535 m a.s.l. RŠ5 is situated just before the confluence with the main stream of the Raška River, 1.7 km from the forth locality, at 540 m a.s.l. Pebble, gravel and stones are the dominant substrate type at all localities.



**Figure 1.** Map with marked localities on the chanell of Raška River

### *Methods*

Macrozoobenthos samples were collected with Surber net, with a catchment area of 300 cm<sup>2</sup> and mesh size of 250 µm. Two samples were taken per locality. The collected material was placed in plastic bottles and preserved with 96% alcohol. The samples were examinad using ZEISS Discovery V8 stereomicroscope in the laboratory of Institute of Zoology, Faculty of Biology. Macroinvertebrates were identified to genus or species level according to following taxonomic keys: Rozkošny1980, Waringer and Graf 1997, Lechthaler and Stockinger 2005, Nilsson 1996, Edington and Hildrew

1981. Zooplankton samples were taken by collecting 30 l of water at each locality using a plastic bucket, and filtering through sieve with 23  $\mu\text{m}$  mesh size. Lentic areas with dense vegetation were chosen. Samples were preserved using 4% formalin and examined under optical microscope Leica Galen III with maximal magnification of 160x. Zooplankton was identified to species or genus level using standard keys for identification (Flosner 1972, Koste 1978). Quantitative samples were analyzed using subsampling technique, where the number of identified species was recalculated to the volume of 1 liter

Saprobity index (S) was calculated according to Pantle-Buck (Pantle and Buck, 1955) method:

$$S = \frac{\sum h \cdot s}{\sum h}$$

h – absolute abundance of individual taxa, s – individual taxa saprobic value. Saprobity values were used from the list of indicator species given by Wegl (Wegl, 1983) for zooplankton and Moog (Moog, 2002) for macrozoobenthos.

#### ***Physical and chemical parameters***

Basic physical and chemical parameters were measured directly in the field and at the laboratory of the Institute for Chemistry, Technology and Metallurgy, Belgrade, Serbia. Measurements of temperature, pH, dissolved oxygen and conductivity were done using water field kit PCE-PHD meter (Germany). Analyses of total phosphorus, orthophosphates (OP) and ionized ammonia ( $\text{NH}_4^+$ ) were performed according to APHA protocols (APHA, 1998).

#### ***Data analysis***

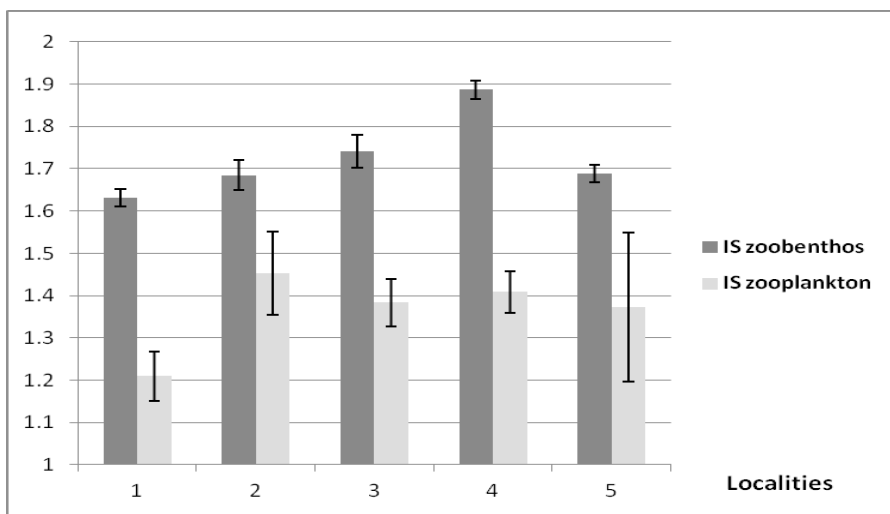
In order to estimate the differences in the saprobity index between localities, t-test was applied using Sigma Stat program (version 2).

### **RESULTS AND DISCUSSION**

A total number of 57 taxa of zoobenthos were found at all studied localities. Of the 57 taxa, 34 were determined to the species level and 21 taxa to the genus level. Oligochaeta and Chironomidae were determined to the class i.e. family level. Aquatic insects were the most diverse group of macroinvertebrates in the Raška River, especially Diptera, with 17 identified taxa (29.82%), followed by Trichoptera (13 taxa, 22.81%), Mollusca (5 taxa, 8.77%), Coleoptera (3 taxa, 5.26%) and Platyhelminthes (1 taxon, 1.75%). The same number of taxa were recorded for Ephemeroptera and Plecoptera (7 taxa, 12.28%) as well as for Hirudinea and *Gammarus* (2 taxa, 3.51%). The diversity of organisms was high at most localities and in all seasons (*Ancylus fluviatilis*, *Valvata cristata*, *Pisidium* sp., *Gammarus balcanicus*, *Gammarus fossarum*, *Baetis* sp., *Ephemerella ignita*, *Ephemerella vulgata*, *Rhyacophila fasciata*, *Rhyacophila tristis*, *Adicella* sp.), but their abundance varied. Taxa sensitive to changes in the aquatic environment (reduced oxygen concentration, high concentration of organic matter) like *Dinocras cephalotes*, *Rhithrogena* sp., *Sericostoma personatum* were found at localities downstream from the trout farm, but were not abundant.

During the research period, a total of 75 zooplankton taxa were found in the Raška River. Rotifers were the most diverse group (65 taxa, 86.67%). Representatives of the zooplankton community also belong to Cladocera (7 taxa, 9.33%), Copepoda (2 taxa, 2.67%) and Ostracoda (1 taxon, 1.33%). The highest number of identified species are among following genera: *Lecane*, *Brachionus*, *Cephalodella* i *Lepadella*. Species common for all studied sites are: *Collurella adriatica*, *Encentrum putorius*, *Euchlanis dilatata*, *Lepadella patella*, *Trichocerca porcellus*, *Mytilina mucronata*, *Alona affinis* and *Alona guttata*.

The composition of zoobenthos communities clearly reflects values of saprobity index. Mean values of the saprobity index along localities ranged between  $1.631 \pm 0.02$  to  $1.88 \pm 0.021$  (Figure 2). The highest values were recorded at localities below the trout farm (RŠ3 and RŠ4), particularly in August (1.91 and 1.94). The saprobity index at RŠ5 was lower and similar to the reference locality RŠ2 (Fig.2). Significant differences in the value of the saprobity index were recorded between fourth and all other localities ( $p < 0.05$ ), i.e. its values clearly indicate a changes in the water quality. The results of the saprobity index show that the water quality along the investigated localities is between the first and second class, i.e. from oligo to beta mesosaprobic.



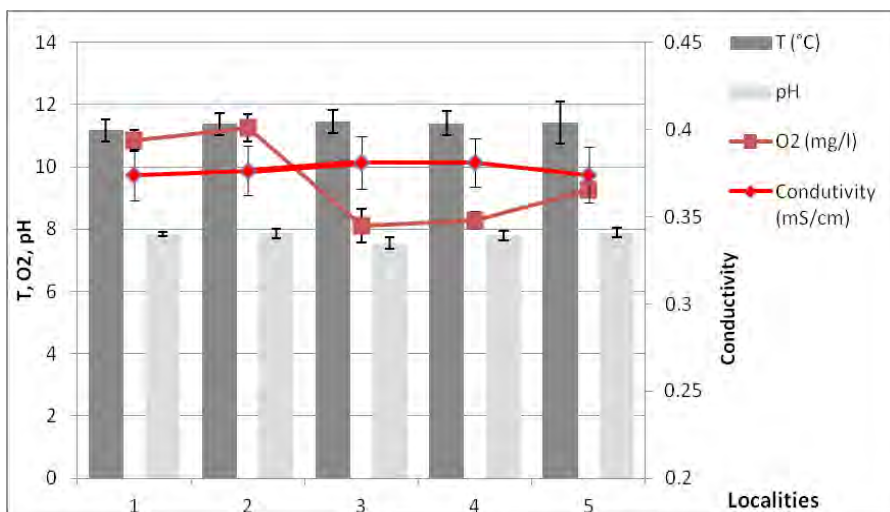
**Figure 2.** Saprobity index values (mean±standard error) for zoobenthos and zooplankton communities

Index of saprobity obtained from the analysis of zooplankton communities indicate a rather different trend. The results of t-test show that the differences of the saprobic index values are almost absent and the only small difference is between first and second locality ( $p=0.049$ ). The highest mean value is recorded at the second locality ( $1.453 \pm 0.098$ ), whereas the lowest values, as in the case of zoobenthos, are observed at the first locality. When using zooplankton organisms, the saprobity index value indicates the first class of water quality along the course, i.e. oligosaprobic.

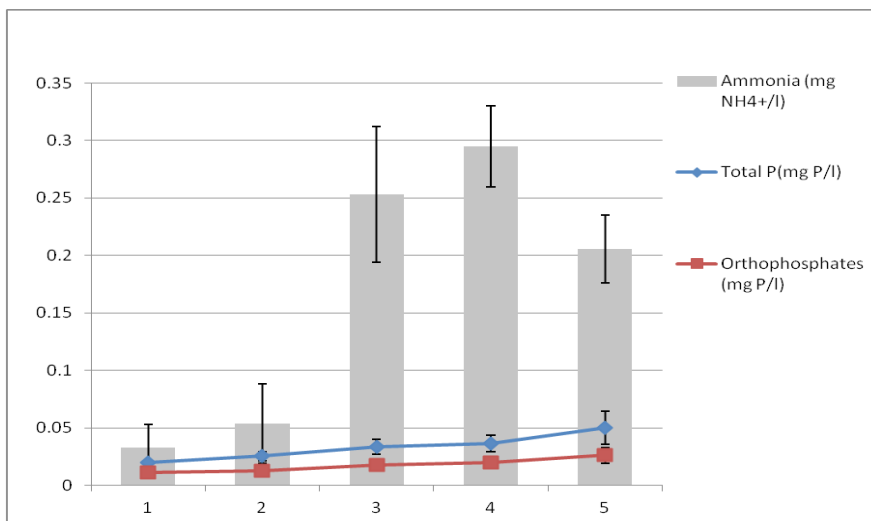
Mean values of basic chemical parameters such as temperature, pH and conductivity, were rather constant at all localities (Figure 3). The exception was a remarkable decrease in the mean concentration of dissolved oxygen at the third locality. This was expected

since RŠ3 is the first locality below the fish farm that regularly discharges water used for trout farming. Concentration of orthophosphates and total phosphorus were below the recommended upper limit (TPECD, 2006) (Figure 4). Downstream from the recipient a slight increase in the concentration of these two parameters was observed. Ammonia concentration varied considerable during the research period (Figure 4). During August and October at RŠ1 and RŠ2 localities values of this parameter were below the limit of detection ( $<0.01$  mg/l), whereas in other months its concentration ranged from 0.01 to 0.05 mg/l. Rapid increase in the concentration of ammonia was noted at the localities below the trout farm. At the localities RŠ3 and RŠ4, the mean values of this parameter were higher than recommended for salmonid waters (Fig. 4)(TPECD, 2006).

By comparing the results obtained from chemical and biological analysis, we can conclude that the values of chemical parameters correlate strongly with the saprobic index of macrozoobenthos. This holds especially for ammonia and oxygen concentration (Figure 3 and 4). Sudden changes in these parameters clearly indicate the influence of the trout farm on the water quality as well as on zoobenthos communities. Oligocaheta, Chironomidae, *Gammarus* and *Simulium* as one of the most tolerant taxa to organic pollution were dominant at RŠ3 locality.



**Figure 3** Mean values ( $\pm$ standard error) of pH, oxygen, temperature and conductivity at studied localities



**Figure 4.** Mean values ( $\pm$ standard error) of total phosphorus, orthophosphates and ammonia at studied localities

Majority of sampled zooplankton organisms belong to the I and II water quality class (Wegl, 1983). The exception is genus *Brachionus*, usually present in waters of second and third class of quality. At the localities downstream from the trout farm, only one species, *Brachionus calyciflorus*, was recorded, while other species of this genus were found at the localities upstream from the trout farm (*B. angularis*, *B. diversicornis*, *B. falcatus*, *B. rubens*). Although 75 taxa of zooplankton were found at the investigated localities, Rotifers were the absolute dominating group. One of the reasons for their prevalence in lotic systems, could be that most of the species found are those adapted for living in interstitial sediments of running waters, especially those with gravel and pebble sediments (Schmid-Araya, 1998). A lot of authors reported high abundance and diversity of Rotifers in streams and rivers of various orders and can constitute from 35 to 85% of the meiofaunal community (Palmer, 1990a,b, 1992; Schmid-Araya, 1997, 1998)

## CONCLUSIONS

Taking into account biological and chemical parameters, water quality of the Raška River was between the first and the second class of the quality.

Results of the saprobiological analysis of zooplankton showed better water quality (oligosaprobic) while macrozoobenthos communities showed that water was betamezosaprobic at all studied localities. This difference was probably due to drift effect in running waters affecting zooplankton communities. Additionally, changes in their populations are more subtle and might need a longer period of investigation to provide comparable results.

Chemical parameters showed correspondence with the macrozoobenthos index of saprobity. The slight increase in total phosphorus and orthophosphates concentrations from the third to the fifth locality, highest ammonia concentration at the third and fourth locality and decline of oxygen concentration, especially at the third locality were followed by an adequate decrease in the saprobity index.

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