COMMUNITY STRUCTURE AND DIVERSITY OF MACROZOOBENTHOS IN QUARRY RIBNICA’S CREEK AS INDICATOR OF SURFACE WATER MANAGEMENT

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Abstract

Benthic macroinvertebrata (macrozoobenthos) are widely used for water quality assessments because they are considered as one of the best indicators of habitat quality in running waters, very conventionally called as „river health“. Benefits of research on macroinvertebrates include the quick assessment of biological resources for conservation purposes and the detection of water pollution. This research was performed in accordance with the EU Water Framework Directive (WFD). AQEM methodology was used for macrozoobenthos specimen collection from six sampling sites along quarry Ribnica’s main hydroecosystems – the Forest creek (quarry’s water supply system) and the estuary of the river Ribnica (Mar-Sep, 2016). Laboratory analyses were carried out for the purpose of separation, counting and final taxonomic identification of organisms. Cluster analysis and diversity index computing were performed using BioDiversity Pro 2.0 software. Our findings suggest the existence of 15 taxa from classes Gastropoda, Hirudinea and Insecta with a total number of 454 sampled individuals. The Insecta class was the most numerous one with respect to the species found, including larvae of the orders Ephemeroptera, Plecoptera, Trichoptera and Diptera. Shannon-Weaver and Simpson diversity indices revealed highly developed communities of aquatic macroinvertebrates at all sampling sites. Water quality along quarry Ribnica based on macrozoobenthos indicator species of saprobity was found to be classified into moderate, low and non to very low polluted (Quality class I-II). Further long-term monitoring studies of macrozoobenthos are necessary in order to keep track of the good ecological status (GES) observed at quarry Ribnica’s aquatic ecosystems.

Key words: macrozoobenthos, biodiversity, water quality, forest creek, river Ribnica

Introduction

Quarry Ribnica is situated on the southern slopes of Ravan Mountain, 6 km northwest of the city Kakanj. The exploitation area in the quarry covers 23.2 ha with mining and excavation activities, while separation processes take place at the Cement Factory Kakanj. No industrial wastewaters are formed during excavation. A small forest creek flows nearby the quarry and right next to the creek, over the industrial site, a water supply reservoir (10-15 m3/day) is placed. The amount of water in the collecting creek can reach up to 20-25 m3/day. Waters from the creek are partially canalized to river Ribnica. Within the industrial site, a concrete precipitator with multiple chambers for acceptance of oily and turbid waters generated by washing machines was built. Waters with the remains of earth, small stones and sand are accepted in the
precipitator chambers, while clarified water is released into a concrete channel. On the way to the river Ribnica, the water passes two settling tanks which are constructed as an additional barrier for the acceptance of particles from turbid waters (Dvokut pro, 2010).

The community of organisms that live on, or in, the bottom of a water body is known as “benthos”. Benthic communities are often used as biological indicators because they can provide information on environmental conditions either due to the sensitivity of single species (indicator species) or because of some general feature that makes them integrate environmental signals over a long period of time. These features are: (i) exposure to chemical contaminants often accumulated in the sediment, (ii) exposure to low dissolved oxygen levels (hypoxia/anoxia) that often occur near the bottom surface due to organic matter degradation, (iii) taxonomic and functional diversity that make them suitable for the detection of different types and levels of stress (Tagliapietra & Sigovini, 2010).

Macrozoobenthos is defined as invertebrate bottom fauna which is retained on a sieve with a mesh size of 1 mm x 1 mm, taking part in several habitats that are protected under the EU Habitats Directive (92/43/EEC): Sandbanks (1110), Estuaries (1130), Mudflats (1140), Lagoons (1150), Shallow inlets and bays (1160), Reefs (1170). Macroinvertebrates, especially reophile species of water insects, show the highest diversity in the creeks (Trožić-Borovac & Ćučuković, 2015). According to the EU Water Framework Directive (WFD), macrozoobenthos is one of the biological quality elements that have to be used to make a classification of the good ecological status (GES) of coastal waters. The generic definition of a GES for benthic invertebrate fauna is: “The level of diversity and abundance of invertebrate taxa is slightly outside the range associated with the type-specific condition” and “Most of the sensitive taxa of the type-specific communities are present” (WFD Annex V) (van der Graph et al., 2009).

The main objective of this research is to determine the qualitative and quantitative composition of benthic macroinvertebrata in order to assess the habitat quality in running waters in relation to the influence of quarrying in quarry Ribnica’s main aquatic ecosystems. Furthermore, this research provides pivotally data on the community structure of macrozoobenthos and ecological status evaluation of the forest creek (quarry Ribnica’s water supply system) and the estuary of river Ribnica, Kakanj, Bosnia and Herzegovina.

Materials and methods

Sample collection, Laboratory Analysis and Taxonomic Identification

In general, macrozoobenthos species were sampled twice annually, in late winter/early spring and in late summer/early autumn (Mar-Sep, 2016). Samples were taken along transects from 6 macrohabitats (Fig. 1A-F) divided into 24 microhabitats.

![Figure 1. Sampling sites along quarry Ribnica creek: (A) L1- Above the base, creek source with water collector; (B) L2- Beneath the base, canalized water source at the foot of the base; (C) L3- Above the precipitator, canalized water from the first depositional system; (D) L4- Beneath the precipitator, canalized water beneath the first depositional system; (E) L5- Canyon, canalized water from the second depositional system; (F) L6- Estuary, mouth of the forest creek in river Ribnica (Authors: Šemir Dorić & Adnan Ćučuković)](image)

Hydro(geo)graphic features of sampling sites are described in Table 1. In the laboratory, organisms were separated from the substratum with forceps and entomological pins, photographed, counted and determined with a binocular loupe / digital microscope and several taxonomic keys [Waringer & Graf (1997), Bole (1969), Elliot et al. (1988), Belfiore (1983), Consiglio (1980), Sansoni (1992),]
Heynes (1977), Moretti (1983), Studemann (1992) and Schmedtje & Kohmann (1992)] to the lowest possible taxonomic category.

Physical and chemical parameters of the forest creek and estuary of the river Ribnica have been taken from a study conducted in previous years (Tab. 2, Dvokut pro, 2010). Temperature and saturation were measured again during sampling with WTW–Multiline F/SET-3 (ATS Abwassertechnik GmbH & Co) and have been used instead of previous measurements.

Cluster Analysis and Diversity Indices

The obtained data from laboratory analyses were processed using the statistic software for ecology and water quality assessment Biodiversity Pro 2.0 (McAleece et al., 1997) and Microsoft Excel™. Three different diversity indices have been used to determine the degree of benthic macroinvertebrata species richness: (1) Simpson Index (D) - a measurement that accounts for the richness and the percent of each species from a biodiversity sample within a local aquatic community, (2) Shannon-Weaver Index (D) - a measurement that takes into account species richness and proportion of each species within the local aquatic community, (3) Saprobic Index (S) - for any given species is the product of abundance $a_i$ and saprobic zone preference $s_i$ expresses the saprobic value $S_i$ for that species. The sum of saprobic values for all the indicator species determined at the sampling point divided by the sum of all the frequency values for the indicator species gives the Saprobic Index which can be calculated from the following formula: $S=\sum_{i=1}^{n}(s_i.a_i) / \sum_{i=1}^{n}(a_i)$. Furthermore, cluster analyses were performed to reveal similarities in composition between individual samples of macroinvertebrates (sampling sites).

Results and Discussion

Qualitative and quantitative analysis of the composition of benthic macroinvertebrates from six sampling sites indicates the existence of 15 taxa with a total number of 454 sampled individuals. In the collected material of the analyzed forest creek and river Ribnica macrozoobenthos organisms from the following classes were found: Gastropoda, Hirudinea and Insecta (Fig. 2A–I). The Insecta class was the most numerous one with respect to the species found, including larvae of the following orders: Ephemeroptera (*Ephemera danica* Müller, 1764, *Ecdyonurus* sp. Eaton, 1868), Plecoptera (*Nemoura* sp. Latreille 1796, *Nemurella picteti* Newman 1853, *Protonemura* sp. Kempny 1898, *Perla* sp. Geoffroy, 1762) Trichoptera (*Allogamus uncutus* Schmid 1955, *Chaetopteryx fusca* Brauer

| Code | Sampling Site | Altitude (m) | Coordinates | Sediment | Coast | Water Level (m) | River Bed (m) |
|------|---------------|--------------|-------------|----------|-------|----------------|---------------|
| L1   | Above the base| 585          | 44° 9′ 29.23″ N 18° 4′ 0.29″ E | Microlital | Low | 0.05 | 0.5 |
| L2   | Beneath the base| 549          | 44° 9′ 22.20″ N 18° 4′ 6.04″ E | Mesolital | Steep | 0.10 | 1 |
| L3   | Above the precipitator | 531      | 44° 9′ 23.03″ N 18° 4′ 8.53″ E | Psamal | Flat | 0.05 | 0.5 |
| L4   | Beneath the precipitator | 502      | 44° 9′ 22.88″ N 18° 4′ 12.53″ E | Fital | Steep | 0.10 | 0.5 |
| L5   | Canyon        | 467          | 44° 9′ 24.35″ N 18° 4′ 15.49″ E | Mesolital | Steep | 0.05 | 2 |
| L6   | Estuary       | 410          | 44° 9′ 22.06″ N 18° 4′ 21.14″ E | Psamal | Flat | 0.5 | 5-7 |

Table 1. Hydro(geo)graphic data of sampling sites in quarry Ribnica
Table 2. Physical and chemical parameters of water at sampling localities in quarry Ribnica creek

| Code | Water temp. °C | pH | Conduc. (μS/20°C) | CaCO₃ hardness (mg/L) | Total water hardness (mg/L) | SO₄ (mg/L) | Mg (mg/L) | Total nitrogen (mg/L N) | O₂ (%) |
|------|----------------|----|------------------|----------------------|----------------------------|------------|----------|------------------------|--------|
| L1   | 14.5           | 7.26 | 445              | 220                  | 242                       | 150.7      | 4.9      | 1.12                   | 79.2   |
| L2   | 15.0           | 7.26 | 387              | 160                  | 234                       | 153.0      | 17.5     | 1.25                   | 80.3   |
| L3   | 16.5           | 7.76 | 460              | 250                  | 274                       | 117.1      | 9.2      | 1.30                   | 79.0   |
| L4   | 14.5           | 7.59 | 236              | 120                  | 134                       | 60.0       | 2.9      | 1.46                   | 82.0   |
| L5   | 14.5           | 7.35 | 282              | 130                  | 148                       | 66.5       | 4.9      | 1.38                   | 80.1   |
| L6   | 15.0           | 7.32 | 309              | 130                  | 152                       | 62.0       | 4.4      | 1.46                   | 84.3   |

*Analysis of physical and chemical water parameters according to Dvokut Pro, 2010. Temperature and saturation were observed during field researches (Mar-Sep, 2016) and average values are showed.

1857, Limnephilidae, Kolenati, 1848, Rhyacophila sp. Denning, 1965, Sericostoma sp. Curtis, 1834) and Diptera (Psychodidae, Williston, 1893) (details are presented in Table 3).

Results of Shannon-Weaver and Simpson diversity index computing suggest highly developed communities of aquatic macroinvertebrates at all sampling sites what causally indicates a high level of water quality. These diversity indices are based on the fact that the ecosystem diversity of species is reducing under the influence of pollution.

According to both indices, the highest diversity of these organisms was found at locality L5 (Shannon-Weaver 0.658; Simpson 0.218) (Fig. 3). Given that this sampling site is very close to the mouth of the creek into the river Ribnica, and beneath two settling tanks, it is significant that these precipitators perform their function very well and thus prevent possible pressure on aquatic ecosystems of the area.

On the other hand, the lowest variety of organisms according to both indices was at locality L6 (Shannon-Weaver 0.301; Simpson 0.467), primarily due to the depth of the riverbed, the flow rate and predators. Also, an enhanced anthropogenic influence could have caused reduced species diversity at this site as well as a set of environmental factors that have caused the development of only certain taxa. This is supported by the fact that the sampled species from this sampling site were significantly larger in comparison to those of other samples. Average Shannon-Weaver and Simpson index values for all six sampling sites were calculated as 0.485 and 0.369, respectively. Cluster analysis, based on the Bray-Curtis index of diversity, indicate the dependence of the composition and

![Figure 2. Biodiversity of macrozoobenthos species found at quarry Ribnica: (A) Gastropoda (snails) - ventral view; (B) Gastropoda (snails) – dorsal view; (C) Hirudinea (leeches); (D) Plecoptera (stoneflies); (E) Trichoptera (caddisflies); (F) Trichoptera (caddisflies); (G) Ephemeroptera (mayflies); (H) Plocoptera (stoneflies); (I) Diptera (true flies) (Authors: Semir Dorić & Adnan Ćučković)]
Table 3. Qualitative and quantitative analysis of macrozoobenthos community at six sampling sites along quarry Ribnica creek

| Taxa                | Species Abundance | L1 | L2 | L3 | L4 | L5 | L6 |
|---------------------|-------------------|----|----|----|----|----|----|
| GASTROPODA          |                   |    |    |    |    |    |    |
| Lymnaea ovata       |                   | 4  | 2  |    |    |    |    |
| Lymnaea sp.         |                   | 36 | 8  | 4  |    |    |    |
| HIRUDINEA           |                   |    |    |    |    |    |    |
| Gen.spec.           |                   | 2  | 4  |    |    |    |    |
| INSECTA             |                   |    |    |    |    |    |    |
| Ephemeroptera       |                   |    |    |    |    |    |    |
| Ephemera danica     |                   |    |    |    |    |    |    |
| Ecdyonurus sp.      |                   |    |    |    |    |    |    |
| Plecoptera          |                   |    |    |    |    |    |    |
| Nemoura sp.         |                   | 28 | 16 |    |    |    |    |
| Nemurella picteti   |                   |    |    |    |    |    |    |
| Protonemura sp.     |                   | 80 | 6  |    |    |    |    |
| Perla sp.           |                   |    |    |    |    |    |    |
| Trichoptera         |                   |    |    |    |    |    |    |
| Allogamus uncatus   |                   | 64 | 70 | 12 |    |    |    |
| Chaetopteryx fusca  |                   |    |    |    |    |    |    |
| Limnephilidae gen.spec. |               | 20 | 20 |    |    |    |    |
| Rhyacophila sp.     |                   |    |    |    |    |    |    |
| Sericostoma sp.     |                   |    |    |    |    |    |    |
| Diptera             |                   |    |    |    |    |    |    |
| Psychodidae         |                   |    |    |    |    |    |    |

*Presence of taxa at sampling sites, Ø Absence of taxa at sampling sites.
distribution of macroinvertebrates in relation to physical and chemical environmental factors. A high degree of similarity of the investigated sites was found (Fig. 4). The cause of this is primarily the length of the watercourse (ca. 1 km). In this area no great diversity of microhabitats nor variations of basic ecological factors were found.

Figure 3. Shannon-Weaver and Simpson diversity indices for six sampling sites along quarry Ribnica creek

The composition of macroinvertebrate communities are most similar at L5 and L6 sampling sites (50% similarity) and the least similar at localities L1 and L5 (5% similarity). With the type of sediment, flow rate, as well as the pH value, the most common species that determined the similarity of sampling sites were *Ephemera danica* Müller, 1764 and *Ecdyonurus sp.* Eaton, 1868 (Ephemeroptera).

Figure 4. Dendrogram of cluster analysis based on the Bray-Curtis index of diversity

Results of Saprobic indexes computing revealed the habitat quality status at six sampling sites in quarry Ribnica. All calculated values are in the range between 1.34 and 1.96 with an average saprobicity value of 1.62. According to this index, moderate pollution was found to be present at sampling site L1, while very low pollution was observed at localities L5 and L6. Localities L2-L4 were found to be under low pollution (Fig. 5). Given that sampling site L1 is next to the road and very close to a tourist resting place, it is important to point out an enhanced anthropogenic influence which directly influenced the development of macrozoobenthos species and their abundance.

Figure 5. Water quality classification along quarry Ribnica creek based on macrozoobenthos indicator species of saprobity

However, water quality assessment based on benthic invertebrate indicator species revealed that the water is generally classified into Quality classes I-II, ranging between oligo-betamesosaprobity and betamesosaprobity.

Conclusions

The bottom fauna of the forest creek and estuary of river Ribnica is composed of 3 groups of macroinvertebrates with 15 determined taxa. The highest diversity is in the order Trichoptera (Fig. 6), with 5 species from two families. The least diverse groups (with only one identified species) are Hirudinea and Diptera. The most important components in formation of the benthocenoses along quarry Ribnica’s aquatic ecosystems are Trichoptera (29.5%) and Plecoptera (26.7%) larvae. Shannon-Weaver and Simpson diversity indices revealed a highly developed community of aquatic macroinvertebrates at all sampling sites. Moderate pollution was found to be present at sampling site L1, while very low pollution was observed at localities L5 and L6. Localities L2-L4 were found to be under low pollution based on indicator species and their saprobic indices. Existing precipitators along the forest creek perform their function very...
well and thus prevent possible pressure on aquatic ecosystems of the area.

Figure 6. Diversity of sampled Trichoptera species and their cases (Authors: Semir Dorić & Adnan Čučuković)

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