## HYDROBIOLOGICAL MONITORING OF TWO RIVERS FROM THE MARITSA RIVER BASIN BASED ON A BIOLOGICAL QUALITY ELEMENT MACROZOOBENTHOS

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#### Abstract

In 2023, an ecological assessment of the state of two rivers, part of the Maritsa river basin in Bulgaria, was carried out. For the study, benthic macroinvertebrate organisms (macrozoobenthos) were collected during the spring season from the Luda Yana River in the area of the village of Popintsi (Panagyurishte Municipality, Pazardzhik Region) and from the Chepelarska River in the area of Katunitsa village (Sadovo Municipality, Plovdiv Region), designated as biotopes. 512 and 712 specimens of benthic macroinvertebrates were collected from the Popintsi biotope and the Katunitsa biotope, respectively. The macroinvertebrate taxa found from the Popintsi biotope are belonging to 12 orders: Amphipoda, Annelida, Coleoptera, Diptera, Ephemeroptera, Gastropoda, Hemiptera, Lepidoptera, Lumbriculida, Odonata, Plecoptera, Ephemeroptera, Gastropoda, Hemiptera, Lumbriculida, Odona

Key words: benthic macroinvertebrates, Bulgaria, Chepelarska River, ecological assessment, Luda Yana River.

## INTRODUCTION

The Maritsa River rises from Rila Mountain and flows into the Aegean Sea, flowing through the territory of three countries. On the Bulgarian territory, the river has a length of 322 km and a catchment area of 21.084 km<sup>2</sup>. The Luda Yana River (74 km) and the Chepelarska River (86 km) are one of the largest tributaries of the Maritsa River (Kiradzhiev, 2013). Both rivers are part of the East Aegean region in Bulgaria. The Luda Yana and Chepelarska Rivers fall into Ecoregion 7 Eastern Balkans. According to the river typology, the Luda Yana River and the Chepelarska River are R3: Mountain type and R5: Semi-mountain type (Belkinova et al., 2013; East Aegean River Basin Directorate, 2018). The rivers falling into the East Aegean basin are subjected to intense anthropogenic pressure from a number of activities, such as mining, discharge of industrial and domestic wastewater. urbanization, alteration of the river bed, extraction of aggregates, and others (East Aegean River Basin Directorate, 2018). The water state of the Luda Yana River is mainly influenced by the activity of enterprises related to the extraction and processing of ores, tailings ponds, and others (Gartsiyanova et al., 2020), located in its upper course. The water quality of the Chepelarska River has deteriorated mainly in the lower reaches of the river, as a result of the activities of industry, mining, tailings ponds, and others (Municipal environmental protection program of Assenovgrad Municipality, 2018-2027). The use. management, distribution, and assessment of water resources in the East Aegean catchment basin was considered by Nikolova et al. (2010); Kolcheva (2016; 2019; 2020); Kolcheva & Ilcheva (2016). The Water Framework Directive aims to achieve good surface water's ecological and chemical status. To assess the ecological status of the water, three groups of elements are monitored: physicochemical, biological, and hydromorphological (Belkinova et al., 2013; Kolcheva et al., 2023). According to the Water Framework Directive, biological

quality elements (BQEs) are leading in the ecological assessment of aquatic ecosystems Benthic (Belkinova et al.. 2013). macroinvertebrate organisms (macrozoobenthos) are good bioindicators for assessing the ecological status of river ecosystems (Varadinova et al., 2022). Scientific studies to assess water quality based on BQE macrozoobenthos from the Luda Yana River were carried out by Vidinova et al. (2008) and Georgieva et al. (2014). The water condition of the Luda Yana River was studied by Gartsiyanova et al. (2020); Gartsiyanova (2021); Gartsivanova et al. (2021); Radeva & Sevmenov (2021); Gartsivanova et al. (2022). There are few studies on the benthic macroinvertebrate fauna from the Chepelarska River (Vidinova et al., 2008; Park et al., 2022; Varadinova et al., 2022).

The aim of the present study is to carry out hydrobiological monitoring of the Luda Yana River and the Chepelarska River (part of the Maritsa River Basin in Bulgaria) based on BQE macrozoobenthos.

#### MATERIALS AND METHODS

In the spring of 2023, samples of benthic macroinvertebrate organisms were collected to carry out an ecological assessment of the water state of the Luda Yana River and the Chepelarska River. The sampling was carried out in the Popintsi village, along the Luda Yana River, and in the Katunitsa village, along the Chepelarska River (Figure 1).

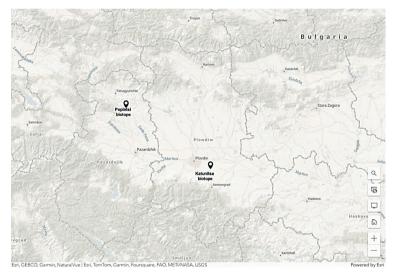


Figure 1. Investigated biotopes from the Luda Yana River and the Chepelarska River (https://www.esri.com/en-us/arcgis/products/arcgis-online/overview)

Sampling of macroinvertebrate organisms is according to Cheshmedjiev et al. (2011); EN ISO 10870:2012; EN 16150:2012; Ordinance H-4 of 14.09.2012; Belkinova et al. (2013). The collected samples were stored in 70% ethyl alcohol for further processing in laboratory conditions. The samples were processed in the laboratory of the Department of Agroecology and Environmental Protection at the Agricultural University - Plovdiv.

Based on an established methodology (Ordinance H-4 of 14.09.2012; Belkinova et

al., 2013; and others), the following metrics were calculated: 1) Taxonomic composition of the benthic invertebrate fauna (total number of taxa; EPT - number of Ephemeroptera, Plecoptera Trichoptera and taxa); 2) Abundance of the benthic macroinvertebrate fauna (% Oligochaeta & Diptera, % Filtering feeders, % EPT taxa and the German trophic index RETI); 3) Saprobity; 4) Indices for species diversity (Species richness index of Margalef (Dmg); Shannon-Weaver species diversity index (H'); Pielou's evenness index

(E); Simpson's dominance index (C) and 5) Integrated indices for the benthic macroinvertebrate fauna (Adapted Biotic Index by Flanagan & Toner, 1972; modified by Clabby & Bowman, 1979; Clabby, 1982).

#### **RESULTS AND DISCUSSIONS**

## Taxonomic composition of the benthic invertebrate fauna

The hydrobiological monitoring was carried out according to the methodology for biomonitoring approved for the European Union and Bulgaria, based on a biological quality element macrozoobenthos. The taxonomic composition of the macrozoobenthos from the two biotopes was determined. In the Popintsi biotope, 23 taxa Nematoda) were (including established, represented by 512 specimens. In the Katunitsa biotope, 20 taxa were found, with a total number of 712 specimens (Table 1). Based on the "total number of taxa", the ecological status of the Luda Yana River (Popintsi) and the Chepelarska River (Katunitsa) was defined as "high" (16+).

Table 1. Taxonomic composition of macroinvertebrate organisms from Popintsi biotope (Luda Yana River) and Katunitsa biotope (Chepelarska River)

TAXON	GENUS	FAMILY	ORDER	вютори			
Acentria ephemerella (Olivier, 1791) [syn. Acentropus niveus (Olivier)]	Acentria Stephens, 1829 Crambidae		Lepidoptera	Katunitsa			
Agapetus sp.	Agapetus Curtis, 1834	Glossosomatidae Trichoptera		Popintsi; Katunitsa			
Anabolia sp.	Anabolia Stephens, 1837 Limnephilidae		Trichoptera	Katunitsa			
Baetis sp.	Baetis Leach, 1815	Baetidae	Ephemeroptera	Katunitsa			
<i>Caenis horaria</i> (Linnaeus, 1758)	Caenis Stephens, 1835	Caenidae Ephemeroptera		Popintsi			
Cataclysta lemnata Linnaeus, 1758	Cataclysta Hübner	Crambidae	Lepidoptera	Popintsi			
<i>Centroptilum luteolum</i> Müller, 1776	Centroptilum Eaton, 1869	9 Baetidae Ephemeroptera		Katunitsa			
Ecdyonurus sp.	Ecdyonurus Eaton, 1868	Heptageniidae	Ephemeroptera	Popintsi; Katunitsa			
<i>Enallagma cyathigerum</i> Charpentier, 1840	Enallagma Selys, 1876	Coenagrionidae Odonata		Popintsi; Katunitsa			
<i>Ephemerella ignita</i> Poda, 1761	Serratella Edmunds, 1959	Ephemerellidae Ephemeroptera		Popintsi; Katunitsa			
Galba truncatula (O.F. Müller, 1774)	Galba Schrank, 1803	03 Lymnaeidae Gastropoda		Popintsi			
Gammarus sp.	Gammarus Fabricius, 1775	5 Gammaridae Amphipoda		Popintsi; Katunitsa			
Gomphus sp.	<i>phus</i> sp. Gomphus Leach, 1815 Gomphidae		Odonata	Popintsi; Katunitsa			
Habrophlebia sp.	Habrophlebia Eaton, 1881	Leptophlebiidae Ephemeroptera		Katunitsa			
Halesus sp.	Halesus Stephens, 1836	sus Stephens, 1836 Limnephilidae Trichopte		Popintsi			
Hirudinea	Annelida		Popintsi; Katunitsa				
Hydropsyche instabilis (Curtis, 1834)	Hydropsyche Pictet, 1834	834 Hydropsychidae Trichoptera		Popintsi			
Hydroporus sp.	Hydroporus Clairville, 1806	Dytiscidae	Coleoptera	Popintsi			
Leuctra nigra (Olivier, 1811)	Leuctra Stephens, 1836	Leuctridae	Plecoptera	Popintsi			
Limnephilus sp.	Limnephilus Leach, 1815	Limnephilidae	Trichoptera	Popintsi			
<i>Nepa cinerea</i> , larva Linnaeus, 1758	Nepa Linnaeus, 1758	Nepidae	Hemiptera	Popintsi; Katunitsa			
Neureclipsis bimaculata (Linnaeus, 1758)	Neureclipsis McLachlan, 1864			Popintsi Katunits			
Notonecta sp.	Notonecta Linnaeus, 1758	Notonectidae	Hemiptera	Popintsi			

TAXON	N GENUS FAMILY		ORDER	BIOTOPE
<i>Physa acuta</i> Draparnaud, 1805	Physella Haldeman, 1842	Physidae	Gastropoda	Katunitsa
TAXON	GENUS	FAMILY	MILY ORDER	
Plectrocnemia conspersa (Curtis, 1834)	Plectrocnemia Stephens, 1836	Polycentropodidae Trichopte		Katunitsa
Simulium sp.	Simulium Latreille, 1802	Simuliidae	Diptera	Popintsi; Katunitsa
<i>Stylodrilus</i> <i>heringianus</i> Claparède, 1862	Stylodrilus Claparède, 1862	Lumbriculidae	briculidae Lumbriculida	
Tabanus sp.	<i>Tabanus</i> Linnaeus, 1758	Tabanidae	Diptera	Popintsi
Taeniopteryx nebulosa (Linnaeus, 1758)	Taeniopteryx Pictet, 1841	Taeniopterygidae Plecoptera		Katunitsa
<i>Tipula</i> sp.	Tipula Linnaeus, 1758	Tipulidae	Tipulidae Diptera	

In Popintsi biotope and Katunitsa biotope, respectively, 9 EPT taxa (39.13% of the established 23 macroinvertebrate taxa) and 10 EPT taxa (50% of the established 20 taxa) were found. Three Ephemeroptera taxa (with 261 specimens), one Plecoptera taxon (with 1 specimen) and five Trichoptera taxa (with 148 specimens) were found in Popintsi biotope. Five Ephemeroptera taxa (with 476 specimens), one Plecoptera taxon (with one specimen), and four Trichoptera taxa (with 148 specimens) were found in the Katunitsa biotope (Figure 2). Regarding the "number of EPT taxa", the ecological status of both rivers was "good".

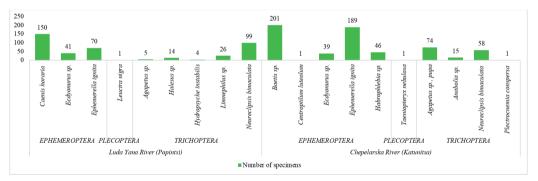


Figure 2. Number of EPT taxa and specimens from Popintsi biotope (Luda Yana River) and Katunitsa biotope (Chepelarska River)

# Abundance of the benthic macroinvertebrate fauna

Oligochaeta taxa have not been identified in the Popintsi biotope. Three Diptera taxa (*Simulium* sp., *Tabanus* sp., and *Tipula* sp.; 7 specimens) were found. In the Katunitsa biotope, one Oligochaeta taxon (*Stylodrilus heringianus*; 14 specimens) and two Diptera taxa (*Simulium* sp. and *Tipula* sp.; 12 specimens) were established. Therefore, the % (Oligochaeta & Diptera) in the Popintsi biotope was 1.37% of the total abundance, and in the Katunitsa biotope -3.65%.

In the Popintsi biotope, one taxon (Simulium sp.; 1 specimen) from the ecological group

"filtering feeders" was found, representing 0.2% of the total abundance. In the Katunitsa biotope, one taxon (*Simulium* sp.; 9 specimens) was also found, i.e., the share of "filtering feeders" is 1.26% of the total abundance.

In Popintsi biotope, 9 Ephemeroptera, Plecoptera, and Trichoptera taxa were found, represented by 410 specimens. Meanwhile, in the Katunitsa biotope, 10 EPT taxa were established, represented by 625 specimens. Therefore, the % EPT taxa in the Popintsi biotope was 80.08% of the total abundance, and in the Katunitsa biotope – 87.78%.

To calculate the RETI trophic index, the trophic groups (SH - shredders; SC - scrapers;

FL - filtering feeders; CL - collectors; DF deposit feeders) of the discovered taxa of benthic macroinvertebrate organisms (including the number of specimens) from the two studied biotopes were defined and presented. In Popintsi biotope, among the trophic groups of benthic macroinvertebrate organisms, the group of deposit feeders (DF; 159 specimens) was represented with the largest number of specimens, followed by the group of shredders (SH; 117 specimens), the group of scrapers (SC; 82 specimens) and the group of filtering feeders (FL; 1 specimen). In the Katunitsa biotope, the group with the largest number of specimens was that of scrapers (SC; 352 specimens), followed by the group of shredders (SH; 256 specimens), the group of deposit feeders (DF; 14 specimens) and the group of filtering feeders (FL; 9 specimens). Taxa from the group of collectors (CL) were not found (Figures 3-4). According to the obtained value of the RETI index for the spring of 2023, the ecological status of the Luda Yana River (Popintsi biotope) was "good" (RETI = 0.548). The Chepelarska River (Katunitsa biotope) was in high ecological condition (RETI = 0.964).

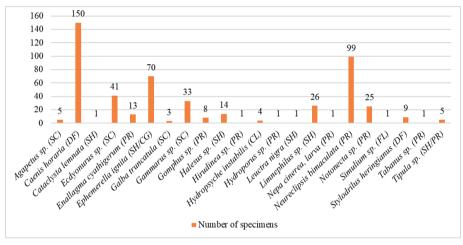


Figure 3. Trophic groups and number of specimens of macroinvertebrate taxa from Popintsi biotope (Luda Yana River)

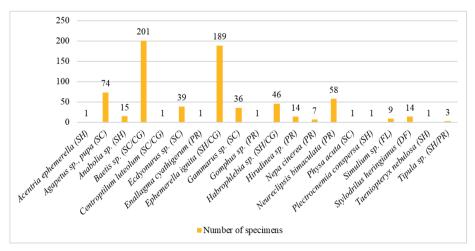


Figure 4. Trophic groups and number of specimens of macroinvertebrate taxa from Katunitsa biotope (Chepelarska River)

#### Saprobity

The detected 22 taxa (without Nematoda) and 20 taxa from Popintsi biotope and Katunitsa biotope, respectively, were found to belong to 8 saprobic groups: xeno-oligosaprobic group (yxeno-β-mesosaprobic 0): group  $(\chi - \beta);$ oligosaprobic group (o); oligo-β-mesosaprobic group  $(o-\beta)$ ; oligo- $\alpha$ - mesosaprobic group (ooligo-polysaprobic group *α*): (0-p): ßmesosaprobic group ( $\beta$ );  $\beta$ - $\alpha$ - mesosaprobic

group ( $\beta$ - $\alpha$ ). In both biotopes, taxa belonging to the following saprobic groups were not found: xenosaprobic group ( $\chi$ );  $\alpha$ -mesosaprobic group ( $\alpha$ ), and polysaprobic group (p). The highest number of taxa and the highest number of specimens, both for Popintsi biotope and Katunitsa biotope, were found for the o- $\beta$ saprobic group (Figure 5).

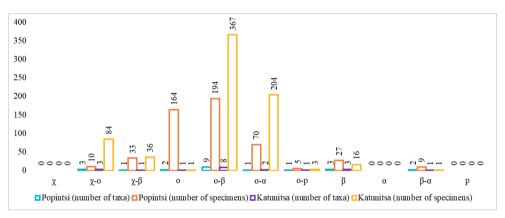


Figure 5. Distribution of detected taxa from Popintsi biotope and Katunitsa biotope by saprobic groups

#### Indices for species diversity

When the species richness index of Margalef has a value above 8, the ecosystem develops optimally (Kirin, 2015), and in the present study, the values of the index are smaller. In both biotopes, the Shannon-Weaver and Pielou index values correspond to  $\beta$ -mesosaprobia. Simpson's index values for both rivers are closer to 0, indicating more favorable conditions (Kirin, 2015) (Table 2).

Table 2. Indices for species diversity

Biotopes	Species richness index of Margalef (Dmg)	Shannon-Weaver species diversity index (H')	Pielou's evenness index (E)	Simpson's dominance index (C)
Popintsi (Luda Yana River)	3.37	2.2	0.711	0.16
Katunitsa (Chepelarska River)	2.89	2.06	0.688	0.179

### Integrated indices for the benthic macroinvertebrate fauna (Adapted Biotic Index)

An Adapted Biotic index (BI) was calculated, for which the detected macroinvertebrate taxa were divided into sensitivity groups: Group A (sensitive), Group B (less sensitive), Group C (relatively tolerant), Group D (tolerant), and Group E (most tolerant). In both biotopes, the detected macroinvertebrate taxa referred to four groups of sensitivity – groups A, B, C, and D. The indicator Group C was represented with the largest number of taxa and the largest number of specimens (Figure 6). In conclusion, the Biotic Index for the Luda Yana River (Popintsi) had a value of 3, corresponding to a moderate ecological condition. At the same time, for the Chepelarska River (Katunitsa), the Biotic Index was equal to 4, i.e., the ecological status is very good.

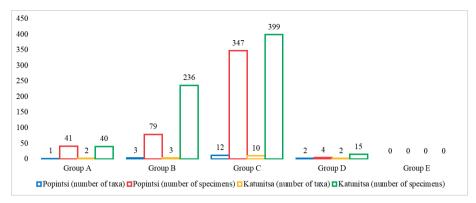


Figure 6. Distribution of detected taxa from Popintsi biotope and Katunitsa biotope by sensitivity groups

#### Discussions

Based on the published data from the East Aegean River Basin Directorate on the surface water state in the period 2014-2021, it was established that in the section of the Luda Yana River from the town of Panagyurishte to the confluence of the Strelchanska Luda Yana River (where Popintsi biotope is located), the ecological status of the water was "bad" and the chemical status - "good". As a result of the report on the water state of the Chepelarska River in the section of Assenovgrad town to the mouth and the Krumovsky collector (where Katunitsa biotope is located) for the period 2014-2020, a "bad" ecological and "bad" chemical status was established. Excesses of Cd and Pb were reported; as well as single excesses of Zn, Mn, and Ni (East Aegean River Basin Directorate, 2018). The current study of the ecological status of the water of the Luda Yana River (Popintsi) and the Chepelarska River (Katunitsa) based BOE on macrozoobenthos shows an improvement of the ecological status of the water of the two rivers in the studied sections in the spring of 2023. It suggests that the better ecological status of the water of the two river ecosystems is due to the higher water level, which is characteristic of the period during which the research was carried out.

## CONCLUSIONS

The hydrobiological monitoring of the freshwater ecosystems of the Luda Yana River (Popintsi biotope) and the Chepelarska River (Katunitsa biotope) was carried out based on the biological quality element macrozoobenthos. According to the applied methodology and the calculated indices, it can be concluded that the ecological status of the Luda Yana River (Popintsi) was moderate ecological condition. In contrast, the ecological status of the Chepelarska River (Katunitsa) was good during the spring when the research was carried out.

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