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Macrozoobenthos as an Indicator of the Ecological State of Mountain Watercourses

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Abstract—Structural characteristics of benthic macroinvertebrate communities in mountain watercourses of different sizes have been studied in the Upper Ob basin. It has been found that species richness, diversity of zoobenthos, and most biotic indices in the background areas increase in the series from the smallest watercourses to large rivers. On the contrary, the values of the above parameters in areas polluted with mercury has proved to decrease, thereby indicating a significant transformation of the benthic communities. Approaches are proposed to select reference indices for assessing the ecological state of watercourses in the basin with regard to the size of the river, the structure of benthic communities, and variability of biotic indices along the background cross sections.

Keywords: macroinvertebrates, ecological state estimation, reference cross sections, heavy metals, mercury

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Development of the biotic concept of control over natural environment implies elaboration and improvement of biological methods used to evaluate the state of aquatic ecosystems. Although the characteristics of macroinvertebrate communities have been recognized and are widely used as promising indicators of the state of river ecosystems, the accuracy and applicability of these methods are limited by local specific features of freshwater fauna. Regional modifications of the known biological methods used to evaluate the state of natural environment do not always provide the desired result, because habitat conditions for macroinvertebrates may differ significantly even within relatively small basins. This leads to changes in the composition and structure of benthic communities, as well as in the biological parameters of water quality calculated on the basis of these characteristics.

A promising approach for resolving these contradictions is to develop a network of reference cross sections and a system of reference parameters based on complex typification of rivers. This approach was promoted by the Water Framework Directive adopted by the EU [1], which stimulated activities aimed at elaborating the principles of selection of reference cross sections, reference parameters, and methods for classification of water quality [2–6].

The purpose of this study was to analyze the distribution of macroinvertebrates in mountain watercourses of the Upper Ob basin in order to develop scientific approaches to the assessment of their ecological state.

MATERIAL AND METHODS

Samples of zoobenthos were collected during the low-water periods of 1989, 1990, 2008, and 2009 in very small rivers (the Yarlyamry, Korumtu, and upper reaches of the Barburgazy), small rivers (lower reaches of the Barburgazy, the Chibitka, Chagan-Uzun, Malaya Siul'ta, and Edigan), and large rivers (four segments of the Chuya and Katun) of the Upper Ob basin. These rivers have similar structure of benthic communities, which was demonstrated previously during typification of zoobenthos in rivers of the Ob basin [7]. Their catchment basins are located mainly in the high and middle (upper layer) mountains, being characterized by either cold-water or moderately cold-water temperature regime, with steep bottom slopes, stony bottoms consisting of boulders and pebbles. The division into size classes was based on the classification of Siberian rivers [8], according to which very small rivers are shorter than 20 km, small rivers are 20–50 km long, medium rivers are 50–200 km long, and large rivers are longer than 200 km.

Bottom samples (boulders and pebbles) were collected with a hydrobiological net (with subsequent calculation of the area of stones by their projection on a plane), then washed through nylon gauze with the mesh size of 350 × 350 μm, animals were singled out and fixated in 70% ethanol. When the constant weight was reached, animals were divided into the taxonomic groups, counted, and weighted with the help of a VT-500 torsion balance. A total of 69 samples of zoobenthos were taken and analyzed.

Table 1. Biotic indices in the background areas of mountain rivers

River	*N*	*B*	*S'*	*H*	TBI	BMWP	ASPT	FBI	EPT
Very small rivers (up to 20 km)									
Korumtu	0.5	0.2	5	2.0	6	36	7.0	2.7	4
Barburgazy (upper reaches)	0.8	0.1	6	2.2	6	9	4.5	5.1	1
Yarlyamry (upstream of AMSW)	0.7	1.1	5	1.5	6	25	6.2	3.2	3
Mean value	0.7	0.5	5.3	1.9	6.0	23.3	5.9	3.7	2.7
Variation coefficient	0.23	1.18	0.11	0.21	0.00	0.58	0.22	0.34	0.57
Small rivers (20–50 km)									
Barburgazy (estuary)	2.1	8.0	12	2.9	7	69	7.0	2.7	7
Chagan-Uzun	0.6	5.2	6	1.5	7	47	6.9	2.8	4
Mal. Siul'ta	10.2	19.7	11	2.4	8	82	7.5	3.1	9
Edigan	5.2	29.3	8	2.3	8	66	7.3	1.9	8
Mean value	4.5	15.5	8.5	2.3	7.5	66.0	7.2	2.6	7.0
Variation coefficient	0.94	0.71	0.48	0.25	0.08	0.22	0.04	0.24	0.31
Large rivers (longer than 200 km)									
Katun near Anos	8.9	3.7	14	2.6	8	63	7.0	3.9	15
Katun near Edigan	1.9	1.7	9	2.4	9	47	7.8	3.5	10
Katun near B. Yaloman	1.4	0.8	5	2.0	7	35	7.0	3.7	4
Katun near Elekmonar	4.0	6.2	11	2.0	9	71	7.9	2.8	6
Mean value	4.1	3.1	9.8	2.3	8.3	54.0	7.4	3.5	8.8
Variation coefficient	0.85	0.77	0.39	0.13	0.12	0.30	0.07	0.06	0.56

N – abundance, 1000 ind./m^2; *B* – biomass, g/m^2; *S'* – average number of species per sample; *H* – Shannon diversity index, bit/ind.; TBI – Trent Biotic Index; BMWP – Biological Monitoring Working Party; ASPT – Average Score Per Taxon; EPT – species number of ephemerans, plecopterans, and trichopterans.

To evaluate the ecological state of mountain watercourses of the Upper Ob basin, we calculated the biotic indices that are most commonly used in the systems of ecological monitoring of rivers in different countries: Trent Biotic Index (Woodiwiss index, TBI); Biological Monitoring Working Party Index (BMWP); Average Score Per Taxon Index (ASPT); Family Biotic Index (FBI); species number of ephemerans, plecopterans, and trichopterans (EPT) [9–13].

Statistical analysis was performed with Statistica 6.0. To estimate the statistical significance of differences, the nonparametric Kruskal-Wallis test (*H*) was used. Differences were considered significant at $p < 0.05$.

RESULTS AND DISCUSSION

Structural characteristics of benthic communities in the background areas of watercourses. The zoobenthos of the studied rivers was represented by 64 species of macroinvertebrates: 10 plecopterans, 14 ephemerans, 15 trichopterans, 18 chironomids, 5 other dipterans, 1 acaridan, and 1 hemipteran.

The benthic communities of the background areas were dominated by amphibiotic insects. *Glossosoma altaicum* Mart. was obviously dominant in most of studied areas. The composition of codominants differed depending on the size of rivers: *Amphinemura borealis* (Morton) in very small rivers, *Brachycentrus americanus* Banks in small rivers, *Arctopsyche ladogensis* (Kolenati) and *Ceratopsyche nevae* (Kolenati) in large rivers. The species richness of the zoobenthos was minimum in very small rivers and maximum in large rivers (Table 1). The low values of species richness and unevenness of spatial distribution of macroinvertebrates resulted in the low values of the Shannon diversity index in very small rivers. An increase in the species richness with distance from headwaters is common to the benthic communities of flowing waters in different regions and, possibly, associated with an increase in heterogeneity of the environment and organic matter content, drift of hydrobionts from upper areas and tributaries, and increase in the abundance of macroinvertebrates [14–17].

In contrast to the species diversity that increased from very small watercourses to large rivers, the maximum abundance and biomass of macroinvertebrates were observed in small rivers (Table 1). The low abundance and biomass in very small rivers may be associated with instability of habitat conditions for hydrobionts in these areas.

Analysis of changes in the biotic indices of the background areas. Most indices used in the systems of biological monitoring of surface waters are based on the phenomenon of decrease in the taxonomic diversity of sensitive hydrobionts with an increase in the pollution level of watercourses. However, the biodiversity may decrease due not only to an increase in anthropogenic pollution, but also changes in the natural biotic and abiotic conditions. To evaluate the possibility of using the selected biotic indices for determining the ecological state of mountain watercourses in Altai, we analyzed the values of these indices in the areas with low anthropogenic load. When investigating the distribution patterns of macroinvertebrates along the longitudinal profile of watercourses, it was revealed that the species richness tended to increase from very small rivers to medium rivers. Thus, the biotic indices were calculated separately for rivers of different sizes.

The values of the biotic indices differed at a statistically significant level depending on river size: for species richness – Kruskal-Wallis test, $H = 13.7$, $p = 0.001$; for Woodiwiss index – $H = 13.15$, $p = 0.001$; for EPT – $H = 17.0$, $p < 0.001$; for BMWP – $H = 14.26$, $p = 0.001$; for Shannon diversity index – $H = 6.79$, $p = 0.033$. Most biotic indices were minimum in small rivers and maximum in large rivers. Due to the low taxonomic richness characteristic of small watercourses, the background values of the Woodiwiss index (TBI) for very small rivers were also low (6) and, according to the State Standard 17.1.3.07-82, corresponded to quality class 3 (moderately polluted waters). The mean values of the BMWP index corresponded to quality class 5 in very small rivers, as well as to class 3 in small and large rivers. Only the ASPT index indicated a high water quality in all studied areas.

One of criteria for applicability of the parameter to evaluation of the ecological state of aquatic objects is its insignificant variability in the reference cross sections. The TBI and ASPT indices proved to be the least variable. The maximum variability was observed for abundance, biomass, and the EPT and BMWP indices, showing that they are poorly applicable to the assessment of the state of watercourses in the Katun basin. The results of analysis show that development of the biotic indices and their regional modifications should involve their gradation by quality classes depending of the river size.

Evaluation of the ecological state of watercourses by structural characteristics of benthic communities. The ecological and geochemical conditions of surface waters in the Katun River basin are formed mainly under the influence of natural factors [18], which provides wide opportunities for selecting reference cross sections. Anthropogenic transformation of the hydrochemical composition of waters is local and associated mainly with the impact of mining industry [19]. To evaluate structural transformations of benthic communities in the polluted mountain watercourses, we analyzed the impact of discharges from the Aktash Mining and Smelting Works (AMSW) located in the middle reaches of the Yarlyamry River, where mercury ore was mined and processed during the study period.

Table 2. Mercury concentration in water (Hg_w, μg/L), suspended matter (Hg_{sm}, μg/g), and bottom sediments (Hg_{ds}, μg/g) in the watercourses of the Katun River, 1990 [21]

River	Hg_w	Hg_{sm}	Hg_{bs}
Yarlyamry, estuary	1.40	177.2	210.80
Chibitka, estuary	0.11	87.2	157.3
Chuya, estuary	0.06	9.1	0.27
Katun, Inya	0.05	2.2	0.27
Edigan, estuary	0.01	3.7	0.03
Katun, Elekmonar	0.02	5.6	0.03
Katun, Edigan	0.02	0.4	0.03

The concentrations of mercury in water, suspended matter, and bottom sediments were the highest in the Yarlyamry River, significantly lower in the Chibitka River, and close to the background level in other rivers of the basin (Table 2). The effect of pollution was traced in the Yarlyamry–Chibitka-Chuya–Katun system.

The zoobenthos of the Yarlyamry River downstream of the AMSW was less diverse in species than in the background areas of rivers of the corresponding size class (very small): the number of species per sample averaged 2.3 ± 0.6. Chironomids (species of the genera *Diamesa*, *Cricotopus*, and *Eukiefferiella*) and simuliids (species of the genus *Simulium*) prevailed both in abundance and in biomass, in contrast to the reference cross sections in very small rivers where *Glossosoma altaicum* and *Amphinemura borealis* dominated. The dominance of simuliids is typical for watercourses that suffer from the impact of mining activities [20], which is probably explained by high resistance of this group of dipterans to heavy metals. During the study period, the clean-water dwellers (trichopterans, ephemerans, and plecopterans) recorded in the cross-section areas, were not found downstream of the AMSW. The species diversity index (Shannon index) of the Yarlyamry River was 0.7–2.0 bit/ind. The high values of the index in certain periods were due to the high evenness of distribution of particular species (mainly chironomids) rather than the high taxonomic diversity.

In the Chibitka River downstream of the confluence with the Yarlyamry River, we found an average of 13.3 ± 0.3 zoobenthos species per sample, which corresponded to the highest values of species richness recorded in the background rivers of this size class. The Shannon diversity index of zoobenthos in the Chibitka River was also relatively high (2.7–2.9 bit/ind.) due to both rich taxonomic composition and high evenness of the abundance distribution of particular species. In the background areas and in the Chibitka River downstream of the confluence with the Yarlyamry River, trichopterans (*Brachycentrus americanus, Glossosoma altaicum*) were dominant in biomass. In contrast to the

Table 3. Values of some biotic indices in the Yarlyamry–Chibitka–Chuya system

River	*H*	TBI	BMWP	ASPT	EPT	FBI
Very small rivers (up to 20 km)						
Background	1.9	6	23.3	5.9	2.7	3.7
Yarlyamry downstream of AMSW	1.4	1	7.0	3.5	0.0	4.2
Small Rivers (20–50 km)						
Background	2.3	7.5	66.0	7.2	7.0	2.4
Chibitka	2.8	6	42.0	7.4	5.0	3.2
Large rivers (longer than 200 km)						
Background	2.3	8.3	54.0	7.4	8.8	3.4
Chuya	2.5	9	53.0	7.7	7.0	2.9
Katun near Inya	2.8	8	55.0	7.5	7.0	3.1

background watercourses, where two or three plecopteran species per sample were found, this group (most sensitive to pollution) was not recorded in the Chibitka River, indicating a poor quality of habitat conditions in the river.

The species diversity of zoobenthos in the Chuya (13.3 ± 0.3 species per sample) and Katun Rivers near the village of Inya (10.5 ± 3.5) corresponded to the same parameters in the background areas of rivers of this size class ($H = 0.18$; $p = 0.9$). The benthic communities of these rivers were characterized by the same taxonomic groups as those in the background areas (including plecopterans). There were no differences in the structure of the complex of dominants that, in both background and test cross sections, was represented by trichopterans such as *Glossosoma altaicum, Arctopsyche ladogensis*, and *Ceratopsyche nevae*. Thus, the conditions for development of benthic communities in the test and reference cross sections were similar.

In order to quantitatively evaluate the degree of anthropogenic transformation of benthic communities in the Yarlyamry–Chibitka–Chuya–Katun system under the impact of discharges from the AMSW, we calculated the indices that are most commonly used in the systems of biological monitoring of surface waters in the European Union, Community of Independent States, and United States. The structure of benthic communities was taken into account when selecting the indices. For instance, rapid current and boulder-pebble bottoms are unfavorable for oligochaetes in mountain rivers. Because of low occurrence frequency and abundance of oligochaetes in the background and polluted areas, it was impossible to use oligochaetan indices. The dominance of Eastern Palearctic elements in the fauna of the studied watercourses accounted for the low proportion of species with the known saprotoxobic valency, which restricted the use of the saprotoxobity index. As a result, the indices based on taxa with the known sensitivity to pollution were singled out. The pollution level for each index was determined in two ways: based on the absolute values in accordance with the quality gradations given for each index by its authors and by ranging the index in relation to the background values. In the latter case, values deviating from the background by no more than 20% were interpreted as high quality; by 20–40%, as good quality; by 40–60%, as satisfactory quality; by 60–80%, as low quality; and by more than 80%, as bad quality. The classes of water quality based on the total number of species and the EPT index were determined only in relation to the background, since there was no absolute parameter to be used as reference in this case.

Most of biotic indices show that the ecological state of watercourses in the Yarlyamry-Chibitka-Chuya-Katun improves with distance from the pollution source (AMSW) (Table 3): TBI, BMWP, and ASPT correspond to water quality classes 5–6 in the Yarlyamry River, classes 1–4 in the Chibitka River, and classes 1–3 in the Chuya and Katun rivers. The use of the other approach (comparison of the biotic indices with background values) allows the quality of water to be classified as high in the Chuya and Katun Rivers, good in the Chibitka River, and satisfactory–low in the Yarlyamry River. It should be noted that there were significant differences between estimates made in the same area using the absolute values of the indices (up to three classes for the Chibitka River), while these differences did not exceed one class when the other method was used.

An important criterion of the possibility to use a bioindication parameter for evaluating the level of anthropogenic impact on aquatic objects is its low variability in the reference cross sections, with significant changes taking place in polluted areas. During the assessment of the ecological state of the most polluted area (the Yarlyamry River downstream of the AMSW), statistically significant differences from the background values were found for all the test indicators: $H = 7.41$, $p = 0.006$ for the Shannon diversity index; $H = 4.93$, $p = 0.026$ for BMWP; $H = 4.93$, $p = 0.026$ for ASPT; $H = 9.85$, $p = 0.002$ for the Woodiwiss index; $H = 7.30$, $p = 0.007$ for EPT; $H = 8.80$, $p = 0.003$ for species richness, which indicates significant transformations in the structure of benthic communities of the impact zone. In the less polluted area (the Chibitka River), where the concentration of mercury in water was lower by an order of magnitude, statistical differences from to the background values were recorded only for Woodiwiss index ($H = 3.76$, $p = 0.05$), ASPT ($H = 13.20$, $p < 0.001$), and BMWP ($H = 4.14$, $p = 0.042$), all least variable in the reference areas. The values of the more variable Shannon diversity and EPT indices in the Chibitka River did not differ from the background values.

On the whole, the minimum variability in the background areas and the maximum changes with distance from the pollution source were recorded for the TBI index, which makes it possible to recommend this index for assessing the ecological state of this type of watercourses. The low variability in the background cross sections and the decrease of the parameters in the most polluted area were recorded for the ASPT index that can be also recommended for this purpose. The FBI index in all areas remained within the limits of classes 1–2 and did not statistically differ between the background and test cross sections. Thus, it is not advisable to use this index for evaluating pollution with heavy metals. Other indices decreased in the most polluted areas but highly varied in the reference cross sections. Therefore, they can be used only in heavily polluted areas where significant changes occur in the structure of communities.

Underrating of water quality in rivers receiving polluted waters and significant variations in the quality classes of nonpolluted river during traditional evaluation by absolute indices are associated with insufficient regard of regional features in the structure of benthic communities. These features in the Altai mountain watercourses include low taxonomic diversity of macrozoobenthos in the region, which leads to underrating of the indices based on the total number of recorded taxonomic groups (such as BMWP). The indices normalized with respect to the number of taxa (such as ASPT) indicate a good ecological state of reference cross sections. Another factor distorting the results of bioindication is the absence of correction for river size in the methods for calculating particular bioindication parameters. The important factor of spatial distribution of macrozoobenthos is the type of bottom sediments, but this aspect is not taken into account in the calculation methods and recommendations for using the biotic indices, which also leads to errors in assessing the ecological state of rivers. These contradictions can be avoided by developing the network of reference cross sections and the system of reference indices based on complex typification of watercourses with regard to their hydromorphological and hydrobiological characteristics.

Therefore, the markedly uneven spatial distribution of macroinvertebrates in rivers of different sizes leads to significant differences in the structural characteristics of zoobenthos and the biotic indices calculated on their basis, as well as makes it necessary to consider the size of the watercourse when selecting the reference cross sections, particularly for rivers within the same basin. An important criterion in selection of bioindicators for evaluating water quality is analysis of their variability in the reference cross sections. The Woodiwiss and ASPT indices can be recommended for assessing the ecological state of rivers with pebble-boulder bottoms in the Upper Ob basin, since these indices show the minimum variability in reference areas and most significant differences in comparisons of reference and impact cross section. Normalization of the indices relative to the background values makes it possible to take into account regional features in the composition and structure of the benthos when determining the quality of waters.

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