EXPERIMENTAL ARTICLES

The Classification and the Monitoring of the State of Mouth Riverine and Lacustrine Ecosystems in Lake Baikal Based on the Composition of Local Microbiocenoses and Their Activity

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Abstract—The paper presents the results of the long-term investigation of microbial communities in the technogenically vulnerable mouth riverine and lacustrine ecosystems of Lake Baikal. The structural and functional parameters of the microbial communities were analyzed from the standpoint of developing destructive processes. The analysis showed that the total number of microorganisms (TNM), the number of saprophytic bacteria (NSB), and bacterial production (BP) were greater in the river-mouth water than in the near-mouth lake water. In the offshore direction, TNM and NSB decreased by a factor of 1.5 to 2, and BP decreased by a factor of 4 to 7. Based on TNM, NSB, and BP data, we classified the Lake Baikal rivers with respect to the degree of the impact of human activities on them. The degrading capability of the riverine microbial communities was found to be such that they degrade daily from tenths of a percent to 3.5% of the total amount of organic compounds polluting the river waters.

Key words: number of microorganisms, bacterial production, rivers, Lake Baikal.

The ecological safety of Lake Baikal depends largely on the purity of the water of its tributaries, most of which flow through industrial zones and carry large amounts of natural and anthropogenic materials. The alluvial cones of the Upper (Verkhnyaya) Angara, Kichera, Rel', Tyya, Tompa, Barguzin, Selenga, and Goloustnaya rivers and the adjacent lake waters are characterized by the transformation and mineralization of the allochthonous material. These processes are mostly implemented by the microbial communities of waters and bottom sediments. This calls for the continuous monitoring of the lake water near the tributary mouths.

The aim of the present work was to study the largest and the most typical (in the degree of anthropogenic impact) aquatic systems near the mouths of the Lake Baikal tributaries.

MATERIALS AND METHODS

Aquatic microbial communities were studied using water samples taken in warm seasons between 1984 and 1999 in large rivers (Selenga, Upper Angara, and Barguzin), rivers polluted with industrial and municipal wastes (Selenga, Tyya, Goloustnaya, Barguzin, Snezhnaya, and some others), and unimpacted rivers (Tompa, Ledyanaya, and Kholodnaya). Water was sampled using either the Sorokin or the Molchanov water sampler in the river-mouth zone-open Baikal ecological system from depths of 0 m (the rivers), 0, 10, and 35 m (the littoral zone of Lake Baikal), and 0, 10, 35, 50, and 100 m (the pelagic zone of Lake Baikal). The total number of microorganisms (TNM) was determined by the direct epifluorescent filter technique [1] using 0.23-µm Synpor membrane filters no. 8 and an MBB-1A microscope (magnification, ×1350; 20 microscope fields examined). The number of saprophytic bacteria (NSB) was determined by the plate technique using fish meal-peptone agar no. 10 (FPA-10). Inoculated agar plates were incubated at 18-20°C for 7-10 days. Bacterial production (BP) was evaluated by the radioisotope method [2] using the coefficient of the heterotrophic carbon dioxide assimilation for Lake Baikal equal to 6.3 [3].

RESULTS

Long-term observations showed that the total number of microorganisms in river waters widely varies (Table 1). The minimal TNM values were detected for the Ledyanaya and Tompa rivers (219 and 399 thousand cells/ml, respectively), which are the least anthropogenically impacted rivers in the Lake Baikal region. The microbiological parameters of the pure water of these rivers were used in this study as the background values. The TNM in the lake water near the tributary

River	Year							max	x	x	$\overline{\mathbf{v}}$	X _r	
	1984	1985	1987	1988	1989	1990	1991	1992	min	min	max	X	$\overline{X_z}$
Selenga	$\frac{1065}{1614}$	$\frac{1250}{700}$	$\frac{600}{600}$	$\frac{4678}{20}$	$\frac{200}{1800}$	$\frac{2700}{2292}$	$\frac{3000}{4000}$	$\frac{4777}{4000}$	$\frac{8.0}{6.7}$	4.2	0.5	$\frac{2509}{2213}$	1.1
Turka	$\frac{1000}{656}$	$\frac{630}{400}$	$\frac{526}{845}$	$\frac{3193}{1800}$	$\frac{2290}{1271}$	$\frac{2100}{1673}$	$\frac{1144}{830}$	$\frac{2534}{1450}$	$\frac{6.0}{5.0}$	3.2	0.5	$\frac{1677}{1116}$	1.5
Barguzin	$\frac{1354}{953}$	$\frac{1450}{256}$	$\frac{718}{330}$	$\frac{4529}{3640}$	$\frac{6650}{3888}$	$\frac{3184}{1000}$	$\frac{1330}{1000}$	$\frac{3704}{3000}$	$\frac{9.2}{15}$	4.0	0.4	$\frac{2865}{1758}$	1.6
Tompa	$\frac{204}{426}$	$\frac{288}{343}$	$\frac{240}{300}$	$\frac{660}{427}$	$\frac{349}{547}$	$\frac{450}{500}$	$\frac{500}{530}$	$\frac{500}{945}$	$\frac{3.2}{3.2}$	2.0	0.6	$\frac{399}{502}$	0.8
Rel'	$\frac{1954}{1067}$	$\frac{600}{500}$	$\frac{300}{700}$	$\frac{566}{830}$	$\frac{229}{615}$	$\frac{466}{1400}$	$\frac{768}{1263}$	$\frac{366}{743}$	$\frac{3.4}{2.8}$	2.9	0.3	$\frac{656}{890}$	0.7
Тууа	$\frac{276}{690}$	$\frac{1432}{458}$	$\frac{377}{285}$	$\frac{712}{1100}$	$\frac{200}{926}$	$\frac{798}{1600}$	$\frac{233}{876}$	$\frac{505}{745}$	$\frac{4.4}{5.6}$	2.8	0.4	$\frac{567}{835}$	0.7
Kichera	$\frac{1397}{1125}$	$\frac{557}{828}$	$\frac{727}{352}$	$\frac{4172}{2000}$	$\frac{100}{1450}$	$\frac{2860}{64}$	$\frac{1350}{430}$	$\frac{2889}{2724}$	$\frac{7.5}{7.7}$	3.4	0.4	$\frac{1869}{1322}$	1.4
Upper Angara	$\frac{1554}{747}$	$\frac{1432}{600}$	$\frac{1500}{729}$	$\frac{4795}{3200}$	$\frac{1797}{1100}$	$\frac{1700}{1675}$	$\frac{1530}{882}$	$\frac{3400}{2518}$	$\frac{3.3}{4.1}$	1.8	0.5	$\frac{2587}{1319}$	2.0
Goloustnaya	$\frac{1800}{1487}$	$\frac{2000}{1300}$	$\frac{2000}{874}$	$\frac{2513}{1500}$	$\frac{1183}{959}$	$\frac{2973}{1586}$	$\frac{3025}{1846}$	$\frac{3957}{1263}$	$\frac{3.3}{1.7}$	2.1	0.6	$\frac{2431}{1346}$	1.8
Ledyanaya	$\frac{241}{200}$	$\frac{167}{200}$	$\frac{100}{475}$	$\frac{390}{342}$	$\frac{190}{319}$	$\frac{257}{320}$	$\frac{225}{180}$	$\frac{182}{159}$	$\frac{3.9}{2.9}$	2.2	0.6	$\frac{219}{274}$	0.8

Table 1. The total number of microorganisms (in thousand cells/ml) in summer in the large rivers of Lake Baikal and in the near-mouth lake water

Note: The numbers that are shown as numerators refer to river waters and the numbers that are shown as denominators refer to the near- X_r

mouth lake water. $\frac{X_r}{X_z}$ is the ratio of the mean value of the numerators to the mean value of the denominators.

mouths was, as a rule, lower than in the river water and decreased still further (by a factor of 1.5 to 2) in the offshore direction. Due to the thermal stratification of the lake water, the TNM in deep water layers was 2 to 3 times lower than in the surface lake water.

The proportion between the numbers of the rivers whose waters were more and less contaminated with microorganisms than the lake water varied from 7:3 to 8:2. The only exception was the year 1989, when this proportion reached 5:5 due to heavy rains during the survey period and the associated high surface runoff.

Interannual variations in the TNM were from 100 to 500 thousand cells/ml for the Ledyanaya and Tompa rivers, from 200 to 800 thousand cells/ml for the Tyya and Rel' rivers, and as low as 1.2 to 4.8 thousand cells/ml for the Goloustnaya and Upper Angara rivers (Tables 1 and 2). Interannual variations in the TNM of the Selenga, Turka, Barguzin, and Kichera rivers reached an order of magnitude. The influence of polluted river waters on the lake water was maximum near the tributary mouths and sufficiently high in the mixing

zone of the river and lake waters (Table 2). At distances greater than 2 km from the mixing zone, the number and the vertical distribution of total microorganisms in summer were typical of the stratified open lake waters, which remain pure and oligotrophic throughout the year due to the coastal circular currents and thermoand pycnoclines isolating the pure deep lake waters from the contaminated littoral waters [4].

Thus, the impact of the river waters on the lacustrine ecosystem of Lake Baikal is restricted to the mixing zones of the lake and river waters, which are located near the river mouths. For instance, the TNM in the water of the Goloustnaya River reached 1 million cells/ml, decreasing to 800 thousand cells/ml in the shallow mixing zone (about 0.5 km from the mouth) and to 500 thousand cells/ml at a distance of 2 km from the mixing zone. Similar data were obtained in June 1992 for the Selenga river. The water of this river contained (4–5) million cells/ml, but 15 km offshore from the river mouth, the total number of microorganisms in the lake water and their vertical distribution corre-

sponded to the average annual values recorded over many years, beginning in 1968 [3].

Microorganisms are delivered to lacustrine microbiocenoses through the dilution, aggregation, and sedimentation of suspended materials. These processes are limited to the 2-km coastal zone [5] except for the Selenga river, whose water influences the microbiological parameters of the lake water even at distances of 5 to 15 km from the mouth. The pelagic zone of Lake Baikal is protected from contamination due to the hydrodynamic sedimentation and assimilation barrier of the coastal ecosystem, which is analogous to the oceanic littoral zone. The stagnant lake waters of this local coastal zone are subject to anthropogenic impact and are characterized by the TNM and NSB values which are anomalously high as compared with those of the pelagic waters of Lake Baikal.

The concentrations of saprophytic bacteria (SB) in the lake water corresponded to their concentrations in the Selenga, Barguzin, Upper Angara, Tompa, Goloustnaya, Turka, Tyya, and Ledyanaya rivers (in the order of decreasing NSB). It should be noted that these rivers are in the same descending order according to the industrial and agricultural human activities on their banks.

The number of saprophytic bacteria, which exhibit a fair growth response to even trace concentrations of organic compounds in water, was at a maximum in the Lake Baikal areas impacted by the severely polluted waters of the Goloustnaya, Selenga, Kichera, Upper Angara, and Barguzin rivers (Table 3). In these areas, the concentration of SB was one to two orders higher than in unimpacted lake areas. The same distribution pattern of saprophytic bacteria in the anthropogenically impacted bodies of water was reported by other authors [6, 7].

The ratio of the maximal NSB to the minimal NSB may serve as an indicator of the ecological stability of the drainage basins of large rivers. The mean number of SB in the river waters was 2 to 3 times greater than in the near-mouth waters of Lake Baikal (Tables 2 and 3) except for the Rel' river, whose waters contained the same number of saprophytic bacteria in the river and in the near-mouth lake zone.

The analysis of the distribution of saprophytic bacteria in the aquatic system river-mouth zone-open lake (2 km distant from the mixing zone of the river and lake waters) showed that the number of saprophytic bacteria in the lake water typically decreased in the offshore direction from the mixing zone, the decrease being especially steep for the Selenga and Barguzin rivers. This, however, was not the case with the Tyya river. Moreover, the number of saprophytic bacteria in the open lake water beyond the mixing zone of waters was greater than in the mouth zone throughout the summer season. This effect may be accounted for by the fact that the fast-running Tyya river carries great amounts of waste organic substances from Severobaikalsk, which

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Table 2. The mean values of the total number of microorganisms and the number of saprophytic bacteria in the large rivers of Lake Baikal and in the near-mouth lake water in the summer–autumn season averaged over the period from 1996 to 1999

River	Depth, m	Total bacteria, thousand cells/ml	Saprophytic bacteria, cells/ml		
Selenga	0	$\frac{1500}{1300}$	$\frac{1890}{868}$		
Upper Angara	0	$\frac{660}{580}$	$\frac{640}{488}$		
Goloustnaya	0	$\frac{100}{890}$	$\frac{452}{170}$		
Turka	0	$\frac{900}{500}$	$\frac{452}{115}$		
Barguzin	0	$\frac{800}{530}$	$\frac{810}{237}$		
Kichera	0	$\frac{640}{400}$	$\frac{230}{156}$		
Tompa	0	$\frac{200}{260}$	$\frac{485}{82}$		
Rel'	0	$\frac{920}{1200}$	$\frac{122}{220}$		
Ledyanaya	0	$\frac{200}{280}$	$\frac{372}{244}$		

Note: The numbers shown as numerators refer to river waters and the numbers shown as denominators refer to the near-mouth lake water.

promote the secondary growth of saprophytic bacteria in the open lake. Indeed, in the case of the reference Ledyanaya river, the number of saprophytic bacteria decreased from hundreds of cells/ml in the river water to tens of cells/ml in the near-mouth lake water and to several cells/ml in the open lake 2 km from the mouth. This indicates a high degree of oligotrophy of both the Ledyanaya River drainage basin and the Lake Baikal area between the Elokhin and Yuzhnyi Kedrovyi capes.

Our calculations showed the existence of a statistically significant correlation (with a coefficient varying from 0.6 to 0.9) between the total number of microorganisms and the number of saprophytic bacteria in the large rivers (Goloustnaya, Selenga, Turka, Barguzin, Kichera, and Upper Angara). At the same time, such a correlation was not observed for the mountain Kholodnaya, Tompa, Ledyanaya, and Tyya rivers.

In general, the activity of microorganisms (bacterial production) in the large rivers was higher than in the near-mouth lake waters, although BP varied widely from year to year (from 3.8 to 70.0 μ g C_{org}/(l day)). Bacterial production in the Ledyanaya river did not

River		1988	1989	1990	1991	1992
Goloustnaya	Ι	1668	559	1717	2565	3360
	II	296	16	732	1051	822
	III	183	3	25	59	152
Selenga	I	2390	2030	3510	3735	6025
	II	2585	1800	1247	2931	2547
	III	236	11	471	70	260
Turka	I	4597	803	1089	1160	1680
	II	1332	419	662	744	713
	III	86	153	304	434	204
Barguzin	I	2187	2170	2570	1560	1489
	II	1777	1360	1600	1130	893
	III	161	220	236	798	77
Tompa	I	225	347	362	263	415
	II	79	219	332	362	290
	III	73	223	11	43	15
Rel'	I	135	622	208	90	43
	II	50	158	128	53	100
	III	22	112	162	26	15
Тууа	Ι	238	310	196	397	505
	II	190	112	205	76	164
	III	145	229	237	110	334
Kichera	Ι	2173	593	699	587	634
	II	166	241	226	228	1256
	III	107	234	277	56	475
Upper Angara	I	1031	760	197	277	706
	II	156	354	347	315	1903
	III	190	507	19	28	158
Ledyanaya	I	176	143	170	100	207
	II	98	41	37	69	13
	III	2	3	2	5	4

Table 3. The distribution of saprophytic bacteria (in cells/ml) in the river and lake water in the summers of 1988 through 1992

Note: I, II, and III refer to, respectively, the river water, the near-mouth lake water, and the open lake water at distances more than 2 km from the mixing zone of river and lake waters.

exceed 3.02 μ g C_{org}/(1 day) throughout the observation period (see figure).

Within 2 km from the mixing zone of waters, bacterial production was 4 to 7 times lower in the lake than in the river water. However, for the open lake waters at distances of more than 2 km from the mixing zone, this regularity was not observed. For instance, bacterial production in the Tyya river was 9.0 μ g C_{org}/(1 day), increasing to 15.0 μ g C_{org}/(1 day) in the open lake at distances more than 2 km from the mixing zone (see figure).

The microbial growth–promoting effect of organic compounds carried by the river waters decreased in the direction from the river to the open lake except for the severely polluted Tyya river, for which the secondary growth of microorganisms was observed (see above). The calculation of the degrading capability of aquatic microbial communities using the experimentally determined assimilation coefficient 0.4 for Lake Baikal [3] showed that these communities may daily degrade from tenths of a percent to 3.5% of the total amount of organic compounds carried by the river waters. The riverine ecosystems of the Rel', Tyya, Kichera, and Upper Angara rivers showed the highest degrading capability. The increased trophic potential of northern Baikal can be explained by the local accumulation of large amounts of organics-rich river waters delivered by the northern tributaries of Lake Baikal.



The mean values of bacterial production in the large rivers of Lake Baikal in summer averaged over a period from 1988 through 1999. Bars 1, 2, and 3 refer to, respectively, the river water, the near-mouth lake water, and the open lake water at distances more than 2 km from the mixing zone of river and lake waters.

A correlation analysis showed that the relationship between the number of saprophytic bacteria and the total bacterial production in the ecosystems studied is complex and variable. The correlation between these parameters was strong (close to a functional dependence) for the Selenga, Turka, Barguzin, Rel', Upper Angara, Ledyanaya, and Goloustnaya rivers (the correlation coefficients r = 0.9, 0.9, 0.7, 0.8, 0.9, 0.9, and 0.8,respectively, at n = 10, $\alpha = 0.01$, and $r_{crit} = 0.7$). For the other rivers, the correlation coefficient was either low or negative, indicating the absence of correlation between the number of saprophytic bacteria and bacterial production. This discrepancy can be explained by the fact that the processes of the assimilation of organic compounds (¹⁴C analysis), the transformation of easily metabolizable substances (the enumeration of saprophytic bacteria), and the metabolism of microbiocenoses (the enumeration of total microorganisms) do not coincide in time.

DISCUSSION

A comparative analysis shows that all methods of bacterial count, including the direct microscopic count and the epifluorescent filter technique, suffer from some disadvantages [8, 9]. Nevertheless, using the method of direct count, we succeeded in elucidating some regularities in the functioning of the riverine and lacustrine ecosystems of Lake Baikal. Based on the total bacterial count estimates, the tributary rivers of Lake Baikal may be divided into three groups. Group I comprises the severely polluted Selenga, Upper Angara, Goloustnaya river, Pokhabikha, and Babkha rivers, which are characterized by the ratio of the total bacterial count in the river water and in the near-mouth lake water equal to 1.1. Group II includes the mildly

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polluted Turka, Barguzin, Kichera, Marituika, and Solzan rivers, which are characterized by the ratio of the total bacterial count in the river water and in the nearmouth lake water varying from 1.3 to 2.3. Group III includes the Tompa, Rel', and Ledyanaya rivers, whose pollution is at a background level. The latter rivers are characterized by the value of the above ratio varying from 0.5 to 0.8.

The ratio of the maximal NSB to the minimal NSB may serve as an indicator of the ecological stability of drainage basins. According to the value of this parameter, 10 large Baikal rivers can be arranged in the following order of descending ecological stability (parenthesized is the ratio under discussion): the Tompa (3.2), Goloustnaya (3.3), Upper Angara (3.3), Rel' (3.4), Ledyanaya (3.9), Tyya (4.4), Turka (6.0), Kichera (7.5), Selenga (8.0), and Barguzin (9.2) rivers. Data on the total bacterial count in the near-mouth lake water can be used for the general ecological characterization of particular rivers.

The fraction of saprophytic bacteria among the total microbial population varied from 0.02 to 0.14%. The high variation in this fraction is most likely due to a high variation in the total microbial population. This fraction cannot be directly used to evaluate the anthropogenic impact on the riverine and lacustrine ecosystems, without differentiating the autochthonous and allochthonous components of the studied parameters. For instance, the differently polluted Tompa and Selenga rivers have the same fractions of saprophytic bacteria, 0.12%. Therefore, this fraction can only be used for the characterization of the internal structure of the riverine and lacustrine microbiocenoses.

The impact of the anthropogenically polluted river waters on the near-mouth lake water results in the fact that bacterial production in the lake water reaches $40 \ \mu g \ C/(1 \ day)$. Similar data were obtained for the alpine oligotrophic Lake Hubsugul in Mongolia [10].

The effect of the organic substances carried by river waters on the microbiological parameters of the lake water persists as far away from the coastal line in the Selenga shallow-water mouth area as 7-15 km, whereas 2-4 km in the case of the other rivers studied. It is this zone where the stable Baikal ecosystem counteracts the adverse effect of polluted river waters.

The ratio of the maximal to the minimal bacterial production in river waters can be used as an indirect characteristic of the ecological stability of drainage basins. This ratio varies within narrower limits (2 to 6 times) than the respective ratios of TNM and NSB, indicating the independence of these parameters. According to the value of the bacterial production ratio, the rivers of Lake Baikal can be arranged in the following order of decreasing ecological stability: Ledyanaya \longrightarrow Kichera \longrightarrow Upper Angara \longrightarrow Tompa \longrightarrow Turka \longrightarrow Selenga \longrightarrow Barguzin \longrightarrow Tyya \longrightarrow Goloustnaya \longrightarrow Rel'. This order corresponds to the increasing anthropogenic load on the particular river or, which is more likely, to the ratio of the total anthropogenic load to the total amount of water in the river.

As an overall conclusion, the data of our long-term ecological and microbiological survey indicate that the ecological state of Lake Baikal and its tributary rivers is stable and corresponds to normal oligotrophic freshwater ecosystems.

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