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Hydrobiological Assessment of the High Mountain Kolsay Lakes (Kungey Alatau, Southeastern Kazakhstan) Ecosystems in Climatic Gradient

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Authors' contributions

This work was carried out in collaboration between all authors. Author EGK designed the study, wrote the protocol and wrote the first draft of the manuscript. Author SMB performed the statistical and bioindicational analysis. Authors SMR and ABM managed the analyses for the study. All authors read and approved the final manuscript.

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ABSTRACT

Aims: The aim of this study is to assess the ecological status of the Kolsay high mountain lakes according to their hydrochemical parameters and structure of phytoplankton communities and reveal the altitude climatic condition impact.

Study Design: We implemented bio-indication and diverse statistical methods, which represent some new approaches in freshwater algal diversity analysis.

Place and Duration of Study: Institute of Evolution, University of Haifa, Israel, Institute of Zoology, Ministry of Education and Science, Almaty, Kazakhstan, between August 2015 and April 2016.

Methodology: We collected 20 samples of phytoplankton in August 2015 from four ultrafresh high mountain Kolsay lakes, located at the altitude of 1829-3170 meters above sea level in the Kungey Alatau, Southeastern Kazakhstan in gradients of climatic and environmental variables that we analyzed. Bio-indication and statistical methods for the ABC Method, Shannon-Weaver index, and WESI index (Water Ecosystem State Index) that indicated some toxic effect on photosynthesis of

algae were used together with Statistica 12.0 and CANOCO Programs for calculating of relationships between environmental and algal communities data among the altitude of the lakes. Results: As a result, 28 species from five taxonomic divisions were identified in phytoplankton communities. According to hydrochemical indices, the lakes were classified as clean, with a slightly increased level of organic pollution in the Middle and Upper Kolsay. The ratio of species in the phytoplankton communities in all the lakes was corresponding to alkaline fresh non-flowing waters. Indication of the indicator species' ratio, functional groups and size structure (the Abundance-Biomass-Comparison method, ABC) of phytoplankton showed that the level of organic pollution decreased in the high-altitude direction. The relationship between the graphic (ABC Method) and the calculation method (Shannon-Weaver index) used for the analysis of the phytoplankton structure is shown. Dynamics of saprobiological and dimensional parameters of phytoplankton links well with the distribution of the recreational load on the lake. The change of biomass of planktonic algae to a greater extent was determined by the dynamics of nitrogen compounds. In general, together with a low level of heavy metals in the water of lakes, the values of WESI index (Water Ecosystem State Index) indicated some toxic effect on photosynthesis of algae in the Lower, Middle Kolsay and the Lake Sary-Bulak. We revealed major variables that considered stimulating or stress factors with helps of statistical programs.

Conclusion: Analysis of hydrochemical and biological data demonstrated that Kolsay mountain lakes are in the early stages of eutrophication. The processes of eutrophication are most evident in the Lower Kolsay. The changes in ecosystems of Kolsay mountain lakes can be caused by the growth of recreational load alongside the altitude related climate change.

Keywords: Mountain lakes; nutrients; heavy metals; phytoplankton; bio-indication; the ABC-method; the Shannon-Weaver index.

1. INTRODUCTION

The water quality management on the watersheds is the goal of modern management, which is impossible without an understanding of the structural changes in the biotic community and identifying the main factors that determine the water quality of aquatic object [1]. Regarding the procedures for assessing the ecological status of natural ecosystems, more and more attention is paid to single-celled algae [2-4]. In order to assess the level of organic pollution of water bodies, different structural indicators of phytoplankton communities are used: the ratio of species - the saprobity indicators [5], the biomass [6], a variety of functional groups and the ratio of diversity to biomass [7,8], the ratio of taxonomic divisions [9,10], size classes of diatoms [11], the value of the average mass of algal cell [12,13].

The graphs of rank distributions of species serve as an integral reflection of community structure [14]. Abundance-Biomass-Comparison (the ABC-Method) has been applied to analyze the structure of benthic [15-17] and planktonic communities [18-21] in the gradient of external factors.

The high mountains Kolsay lakes are located in the Kungey Alatau Mountains [South-Eastern

Kazakhstan] within the territory of the National Natural Park, established in 2007. The lakes are remote from the agricultural and industrial areas. Anthropogenic impact on their ecosystems is associated with recreational load and acclimatization activities. Due to the inaccessibility, the Kolsay Lakes are poorly explored. Fragmentary information is only available on zooplankton and hydrochemical parameters of the two lower lakes [21-23].

Mountain cold-water lakes are more sensitive to external factors compared to lowland reservoirs. Significant changes in hydrocenosis of mountain lakes can occur even in the absence of direct human impact [24]. In order to preserve the mountain lakes as a specially protected object, the methods for capturing the changes on the early stages are needed. The aim of this study is to assess the ecological status of the Kolsay mountain lakes according to their hydrochemical parameters and structure of phytoplankton communities.

2. MATERIALS AND METHODS

Complex studies of the Lower, Middle, and Upper Kolsay Lakes and Sary-Bulak Lake (Fig. 1) were carried out in August 2015. In the field environment, the temperature, pH and electrical conductivity of the surface water layers were measured by means of HANNA HI 98129 devices. Water transparency was measured by Secchi disk. On the each lake, a sample of 1 liter of phytoplankton was taken. Each sample consisted of subprobe selected in three to five different parts of the lake. Subprobes were mixed, and then one integrated sample of required volume was selected. The water samples to determine the mineralization and chemical composition, the content of easily oxidized organic matter, nutrients and heavy metals were collected using the same principle.

The conventional methods of chemical analysis of water were used [25,26]. All water samples were analyzed in three-four replications. Determination of heavy metals in water was performed by mass-spectrographic method with inductively coupled plasma using the Agilent 7500A manufactured by the Agilent Technologies, USA (ST RK ISO 17294-2-2006). Assessment of the ecological state of the lakes on the hydrochemical parameters were carried out according to [6]. Data on the content of nutrients and heavy metals were compared to the maximum permissible concentrations of substances for fishery waters (MPC) [27,28].

For the processing of phytoplankton samples, the settling method was used [29]. Species identification of planktonic algae was performed by using determinants for relevant divisions [30-35]. Shannon-Weaver index was calculated on Primer 5 program through the logarithm with the base 2 in two versions - on the share in the total number of species [bit/individual] and on biomass [bit/mg] [36]. Curves on domination were calculated using the same program as the Abundance-Biomass-Comparison method (ABC) [14] which we implemented earlier for phytoplankton [18-21]. W-statistic of Clarke was calculated automatically [37]. Its value indicates the position of the biomass in respect to the curve the abundance. A positive W value indicates that the biomass curve is higher than abundance curve, and vice versa. The average mass of the algal cell was found as the ratio between the total biomass and the total abundance of phytoplankton.



Fig. 1. Map – location scheme of the Kolsay lakes

The ecological characteristics of algal species were obtained from the database compiled for freshwater algae of the world from multiple analyses of algal biodiversity by S.S. Barinova et al. [3], according to substrate preference, temperature, oxygenation, pH, salinity, organic enrichments, N-uptake metabolism, and trophic states. The ecological groups were separately assessed according to their significance for bioindications. Species that respond predictably to environmental conditions were used as bioindicators for particular variables of aquatic ecosystems, the dynamics of which are related to environmental changes. The statistical methods used were those recommended by Heywood [38] for the development of taxonomic studies, namely the CANOCO program [39] for canonical correspondence analysis (CCA) [40,41], the GRAPHS program [42] for Principal Component Analysis (PCA) and comparative floristics. Surface plots for biological and environmental variables relationship analysis were constructed by using the Statistics 12 program.

The correspondence between the classificational indicators of ecosystem according to the biogenic elements and those of saprobity indices was conducted through calculating the index of the ecosystem state (WESI, Water Ecosystem State Index) as the quotient from the classification rank on biota divided by the classification rank on the environment. The WESI index ranges from 0 to 9. If it is less than one, then the ecosystem is exposed to the toxic effects; if equal to or greater than 1 – the self-purification is not suppressed [3,43].

3. RESULTS

3.1 General Characteristics of the Study Region

The Kolsay lakes are located in the Kungey Alatau Mountains (South-Eastern Kazakhstan) at the altitude from 1829 up to 3170 m a.s.l. Three lakes (Lower, Middle and Upper Kolsay) are flow through, canyon type, deep-water, small in surface area, with high water transparency (Table 1). Originating in the northern and western slopes of the mountains, the river of the same name flows through the lakes. Two more small rivers fall into the Middle Kolsay. The Sary-Bulak Lake is shallow and located in the alpine zone. It is supplied by groundwater and precipitation. A small stream is flowing outflows from the lake and flows into the Kolsay River. Water transparency is to the bottom. The bottom of the lake is overgrown with Charophyta algae.

Lower, Middle and Upper Kolsay lakes are coldwater. The temperature of water decreases with altitude. The water temperature in Sary-Bulak Lake is higher than in the Middle and Upper Kolsay. In August 2015, immediately after the snowfall, its water temperature reached 12.7°C. In 2002 and 2006, the water was heated to a temperature 18-20°C [21]. The water in all lakes is alkaline, with a maximum pH in the Sary-Bulak. Lakes are ultrafresh (Table 2). Mineralization of water decreases in the highaltitude direction. Water is of the carbonate class of calcium group, the second type, very soft. There was small amount of readily oxidized organic matter and nutrients in the water of all lakes. The amount of nitrite in the water decreased in the direction from the lower to the upper lake. Nitrate content varied within a relatively small range. In the Sary-Bulak Lake, the nitrates were not recognized. Ammonium ions were absent in the water of the Lower Kolsay, and in other lakes the concentration changed insignificantly. Phosphorus was not in water.

The content of heavy metals in water, with the exception of copper, were one or two orders lower than the established permissible concentrations of substances [27,28]. The concentration of lead, copper, zinc, nickel and chromium in water decreased from Lower to Upper Kolsay. In the Sary-Bulak Lake, the content of almost all heavy metals were higher than in the downstream lakes.

Table 1. Hydro physical characteristics of Kolsay Lakes, August 2015

Variable	Lower Kolsay	Middle Kolsay	Upper Kolsay	Kolsay Sary-Bulak
Altitude, m above sea level (a.s.l.)	1829	2242	2642	3170
Surface area, km ²	0.58	0.67	0.07	0.02
Maximum depth, m	36.6	54.0	25.0	2.5
Temperature, °C	13.7	12.1	10.1	12.7
pH	8.40	8.62	8.27	9.60

Variable	Units	Lower Kolsay	Middle Kolsay	Upper Kolsay	Kolsay
			-	,	Sary-Bulak
Hardness	mg Eq. dm⁻³	1.4	1.2	1.1	0.25
Deep	m	36.6	54.0	25.0	2.5
Secchi	m	9.0	9.0	9.0	2.5
TDS	mg dm⁻³ ୁ	123.9	116.0	102.2	26.6
BOD	mgO dm⁻³	1.36	1.2	1.2	2.14
NO ₂	mg dm ³	0.028	0.03	0.02	0.011
N-NO ₂	mg dm ً	0.008	0.009	0.006	0.003
NO ₃	mg dm ື	0.688	0.344	0.516	0.0
N-NO ₃	mg dm ຼຶ	0.155	0.078	0.117	0.0
NH_4	mg dm ຼຶ	0.0	0.2	0.15	0.22
N-NH ₄	mg dm ຼຶ	0.0	0.156	0.117	0.171
PO ₄	mg dm ຼຶ	0.0	0.0	0.0	0.0
Fe	mg dm ຼຶ	0.140	0.164	0.044	0.440
Si	mg dm ຼຶ	4.2	6.3	4.4	1.0
Mn	mg dm ີ	0.0165	0.008	0.008	0.010
Pb	mg dm ္ရွိ	0.00014	0.00009	0.0001	0.0005
Cu	mg dm ັ	0.0038	0.0031	0.0026	0.0055
Zn	mg dm ္ရွိ	0.0009	0.0010	0.0002	0.0016
Cd	mg dm ္ရွိ	0.00045	0.0004	0.0006	0.0004
Ni	mg dm ္ရွိ	0.0018	0.0017	0.0014	0.0027
Cr	mg dm⁻°	0.0008	0.0007	0.00065	0.0005
No of Species		15	8	10	6
Cyanobacteria		3	0	0	0
Bacillariophyta		7	8	8	1
Charophyta		0	0	0	1
Chlorophyta		3	0	2	4
Euglenophyta	2	2	0	0	0
Abundance	mln. cells m ^{-s}	510.2	75.0	110.1	11.7
Biovolume	mg m⁻°	225.7	450.7	477.3	78.3
Shannon Ab	bit/individual	2.16	2.38	2.67	2.24
Shannon Bi	bit/mg	2.61	1.94	2.25	1.14
Average cell mass	×10 ⁻ °, mg	0.442	6.009	4.335	6.692
Average W		-0.077	0.127	0.136	0.610
Index Saprobity S		1.51	1.12	0.99	0.76
Index WESI		0.50	0.67	1.00	0.50

3.2 Brief Description of the Lakes Phytoplankton

Altogether 28 species of phytoplankton were identified in Kolsay lakes (Table 3). The major number of them was Bacillariophyta. The highest algal diversity in Division level was recorded in the Middle and Upper Kolsay. Bluegreen algae were found only in the Lower Kolsay.

Phytoplankton in Lower and Middle Kolsay lakes was close in species composition (Fig. 2a). The algae communities of Upper Kolsay and Sary-Bulak were distinguished into separate groups. Similar results were obtained when using PCA analysis (Fig. 2b). The number of phytoplankton cells varied by orders of values, with the maximum index value in the Lower Kolsay and its tendency to decrease with altitude (Table 2). In the Lower Kolsay Lake, blue-green algae dominated up to 90% of the total abundance. Dominant complex included Planktolyngbya contorta (20.6%), P. limnetica (36.3%), and Geitlerinema amphibium (33.3%). Only diatoms were represented in the Middle Kolsav phytoplankton. Diatoms (69.7%) dominated numerically, and green algae (30.3%) sub-dominated in the Upper Kolsay. In these two lakes, Lindavia comta, C. meneghiniana, C. planctonica made the largest contribution formation to the of the community composition (9.1-37.7%). The dominant species complex in the Upper Kolsay also included *Sphaerocystis planctonica* (24.3%) from the green algae. Green algae formed up to 85.5% and diatoms up to 14.5% of abundance in phytoplankton of the Sary-Bulak Lake. The most numerous were diatoms *Lindavia comta* (14.5%), green algae *Closteriopsis longissima* (14.5%), *Monoraphidium contortum* (28.2%), *M. obtusum* (14.5%), as well as *Spirogyra* sp. (28.2%) from Charophyta Division.

The phytoplankton biomass was highest in the Middle and Upper Kolsay lakes. The biomass in the Sarv-Bulak Lake was shaped with 69.7-100.0% by diatoms. The euglenoids were subdominants in the Lower Kolsay Lake making up to 22.9% of the phytoplankton biomass. The Sary-Bulak Lake phytoplankton biomass was represented by green algae, which made the main contribution (81.6%), with diatoms (18.4%) as sub-dominants. The diatoms Cyclotella meneghiniana (20.0-35.4%) and C. planctonica (27.2–38.6 %) occupied the dominant position on biomass in the three lower lakes. The dominant complex in Lower Kolsay was included Trachelomonas intermedia (15.5%). Lindavia comta played a significant role in the formation of phytoplankton biomass of Middle, Upper Kolsay Sary-Bulak (18.4–34.4%) lakes. and The filamentous algae Spirogyra sp. made the largest contribution (74.5%) to the formation of the Sary-Bulak Lake phytoplankton biomass.

Shannon-Weaver index was characterized by the moderate level of the diversity of the algal communities of the lakes (Table 2). The phytoplankton average cell mass was enlarged with increasing altitude. The species richness of phytoplankton communities grew up with the nitrates increase, but not with water mineralization (Fig. 3a), and correlated with the increase in abundance (Fig. 3b) as well. The number of diatom species in phytoplankton grew up with increasing amounts of nitric nitrogen and was highest in smallspecies communities (Fig. 3c), subject to water silicon saturation (Fig. 3d).

3.3 Assessment of the Ecological State of the Lakes Ecosystem by Hydrophysical and Hydrochemical Indicators

Concentrations of heavy metals in the Kolsay lakes water were insignificant, which is typical for the mountain reservoirs of South-Eastern Kazakhstan [48,49]. Most of the analyzed parameters of the environment did not exceed the limits for extremely pure and clean water (I–II Class, Rank 1–3), except for nitrite [6] (Table 4). The value of nitrites corresponded to Water Class III – satisfactory purity in Lower, Middle and Upper Kolsay lakes, and to the level of pure water (Class I) in the Sary-Bulak Lake.

All lakes have alkaline water (Table 2). According to the classification [6], such pH values are characteristic of moderately polluted (three lower lakes) and extremely polluted waters (Sary-Bulak Lake). Evidently, this classification is cannot be used for assessing the ecological status of the Kolsay lakes. High water alkalinity in most water bodies of Kazakhstan is natural and not related to anthropogenic pollution. The pH values higher than 8.4 were observed even in the spring waters of Kazakhstan [50].



Fig. 2. Dendrite showing the similarities in composition of phytoplankton species of the Kolsay lakes (a) and Principal Component Analysis (PCA) (b), August 2015

Таха	Code	LK	MK	UK	KS	Hab	Т	Reo	рН	Hal	D	S	Sap	Tro	Aut- Het
Cyanobacteria															
Geitlerinema amphibium (C.Agardh ex Gomont)	GeiAmp	1	0	0	0	P-B,S	-	st-str	-	hl	-	2.4	o-a	m	-
Anagnostidis															
Planktolyngbya contorta (Lemmermann)	PlaCon	1	0	0	0	Р	-		-		-	-	-	me	-
Anagnostidis & Komárek															
Planktolyngbya limnetica (Lemmermann)	PlaLim	1	0	0	0	P-B,S	-	st-str	-	hl	-	1.5	o-b	-	-
Komárková-Legnerová & Cronberg															
Bacillariophyta															
Achnanthidium minutissimum (Kützing) Czarnecki	AchMin	1	1	1	0	P-B	eterm	-	ind	-	-	1.5	o-b	me	-
Cocconeis placentula Ehrenberg	CocPla	0	1	1	0	P-B	temp	st-str	alf	i	es	1.3	o-b	е	ate
Cyclotella meneghiniana Kützing	CycMen	1	1	1	0	P-B	temp	st	alf	hl	sp	2.8	а	е	hne
Cyclotella planctonica Brunnthaler	CycPla	1	1	1	0	Р	-	-	ind	i	-	-	-	-	-
Cymbella parva (W.Smith) Kirchner	CymPar	1	0	0	0	B	-	-	ind		-	-	b	o-m	-
Diatoma vulgaris Bory	DiaVul	0	1	0	0	P-B	-	st-str	ind	i	SX	2.2	b-a	me	ate
Encyonema minutum (Hilse) D.G.Mann	EncMin	1	0	0	0	В	-	st-str	ind	I	SX	1.2	X-0	ot	-
Fragilaria acus (Kützing) Lange-Bertalot	FraAcu	1	0	0	0	-	-	-		-	-	1.8	b .	-	-
Gomphonema longiceps Ehrenberg	GomLon	0	0	1	0	В	-	str	ind	1	es	1.4	o-b	-	-
Gomphonema olivaceum (Hornemann) Brébisson	GomOli	1	0	1	0	В	-	st-str	alt	1	es	1./	b-a	е	ate
Gomphonema truncatum Enrenberg	Gom I ru	0	1	0	0	P-B	-	st-str	ind	1	es	1.4	b	me	ats
Lindavia comta (Kutzing) Nakov, Gullory, Julius,	LinCom	0	1	1	1	Р	-	st	alf	I	SX	1.2	D-0	-	-
I neriot & Alverson	Neuclin	0	4	~	0	Б		-1 -1-	- 14			<u>م ح</u>			-1-
Navicula cincta (Enrenberg) Raits		0	1	0	0	В	warm	st-str	air	1	es	0.5	X-0	me	ate
l'etracyclus lacustris Raits	TetLacu	0	0	1	0	В	-	at atr	act	1	.	0.4	0	-	-
Planothidum lanceolatum (Brebisson ex Kutzing)	PlaLan	0	I	U	0	Р-В	wann	รเ-รแ	ina	I	SX	1.0	D	0-111	-
Lange-Bertalot															
Trachalomanas hispida (Portu) E Stoin	Trollio	1	0	0	Δ	DD	otorm	ot otr		;		22	h		
Trachelomonas intermedia P.A. Dangoard	Traint	1	0	0	0	г-D D D	otorm	5เ-รแ	-	1	-	2.3	b	-	-
Chlorophyta	nann	I	U	U	U	r"-D	eleilli	-	-	I	-	۷.۷	U	-	-
Chlorolohion braunii (Nägeli) Komárek	ChlBra	1	٥	0	Ο	D_R	_	et	_	_	_	15	o_h	_	_
Closterionsis longissima (Lemmermann)	Chllon	1	0	0	1	D	-	ol et_etr	-	-	-	1.0	0-0	-	-
Bacillariophyta Achnanthidium minutissimum (Kützing) Czarnecki Cocconeis placentula Ehrenberg Cyclotella meneghiniana Kützing Cyclotella planctonica Brunnthaler Cymbella parva (W.Smith) Kirchner Diatoma vulgaris Bory Encyonema minutum (Hilse) D.G.Mann Fragilaria acus (Kützing) Lange-Bertalot Gomphonema longiceps Ehrenberg Gomphonema olivaceum (Hornemann) Brébisson Gomphonema truncatum Ehrenberg Lindavia comta (Kützing) Nakov, Gullory, Julius, Theriot & Alverson Navicula cincta (Ehrenberg) Ralfs Tetracyclus lacustris Ralfs Planothidium lanceolatum (Brébisson ex Kützing) Lange-Bertalot Euglenophyta Trachelomonas hispida (Perty) F.Stein Trachelomonas intermedia P.A. Dangeard Chlorophyta Chlorolobion braunii (Nägeli) Komárek Closteriopsis longissima (Lemmermann)	AchMin CocPla CycMen CycPla CymPar DiaVul EncMin FraAcu GomLon GomOli GomTru LinCom NavCin TetLacu PlaLan TraHis TraHis TraInt ChIBra ChILon	$ \begin{array}{c} 1 \\ 0 \\ 1 \\ 1 \\ 0 \\ 1 \\ 0 \\ 0 \\ 0 \\ 0 \\ 1 \\ 1 \\ 1 \\ 1 \\ 1 \\ 1 \\ 1 \\ 1 \\ 1 \\ 1$	1 1 1 0 0 0 0 1 1 1 0 1 0 0 0 0 0 0 0	$ \begin{array}{c} 1 \\ 1 \\ 1 \\ 0 \\ 0 \\ 0 \\ 1 \\ 0 \\ 1 \\ 0 \\ 0 \\ 0 \\ 0 \\ 0 \\ 0 \\ 0 \\ 0 \\ 0 \\ 0$	0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0	P-B P-B P-B B P-B B P-B P-B P-B P-B P-B	eterm temp - - - - - - - warm - warm - warm - - warm - - -	- st-str st-str st-str st-str st-str st-str st-str st-str st-str st-str st-str st-str	ind alf ind ind alf ind alf alf acf ind - -	- 	- es sp - sx es es sx es sx es sx - -	1.5 1.3 2.8 - 2.2 1.2 1.2 1.8 1.4 1.7 1.4 1.2 0.5 0.4 1.6 2.3 2.2 1.5 1.8	o-b o-b a - b b-a b b-a b b-a b b-o x-o o b b b o-b	me e - o-m me - e me - me - o-m - -	- ate - ate ate ate - - ate - - - - - - - - -

Table 3. Taxonomy and ecology of the algal communities in the Kolsay lakes

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Таха	Code	LK	MK	UK	KS	Hab	Т	Reo	рН	Hal	D	S	Sap	Tro	Aut- Het
Lemmermann															
Crucigenia quadrata Morren	CruQua	0	0	1	0	P-B	-	st-str	acf	i		1.9	b-o	-	-
Monoraphidium contortum (Thuret) Komárková-	MonCon	0	0	0	1	Ρ	-	-	-	i				-	-
Legnerová	-														
Monoraphidium griffithii (Berkeley) Komárková- Legnerová	MonGri	1	0	0	0	Р	-	-	-	i		2.2	b	-	-
Monoraphidium obtusum (Korshikov) Komárková- Legnerová	MonObt	0	0	0	1	Р	-	-	-	i		2.2	b	-	-
Sphaerocystis planctonica (Korshikov) Bourrelly Charophyta	SphPla	0	0	1	0	Р	-	-	-	i	-	-	-	-	-
Spirogyra sp.	Spirog	0	0	0	1	В	-	-	-	-	-	-	-	-	-

Note: LK – Lower Kolsay, MK – Middle Kolsay, UK – Upper Kolsay, KS – Kolsay Sary-Bulak. Note: Substrate preferences (Habitat): P – planktonic, P-B – plankto-benthic, B – benthic, S – soil. Temperature preferences (Temp): temp – temperate temperature, eterm – eurythermic, warm – warm-water. Oxygenation and streaming (Reo): st – standing water, str – streaming water, st-str – low streaming water. Halobity degree according Hustedt [44] (Hal): i – oligohalobes-indifferent, hl – halophiles. Acidity (pH) degree according Hustedt [45]: alf – alkaliphiles, ind – indifferents; acf – acidophiles. Organic pollution indicators according to Watanabe et al. [46] (D): sx – saproxenes, es – eurysaprobes, sp – saprophiles. Species-specific Index of Saprobity (S). Self-purification zone preferences (Sap): x – xenosaprob; o – oligosaprob; o-b – oligo-betamesosaprob; o-a – oligo-alpha-mesosaprob; b – beta-mesosaprob; b-a – beta-alpha-mesosaprob; a – alpha-mesosaprob. Nitrogen uptake metabolism (Aut-Het) [47]: ats – nitrogen-autotrophic taxa, tolerating very small concentrations of organically bound nitrogen; ate – nitrogen-autotrophic taxa, tolerating elevated concentrations of organically bound nitrogen; hne – facultatively nitrogen-heterotrophic taxa, needing periodically elevated concentrations of organically bound nitrogen. Trophic state (Tro) [47]: ot – oligotraphentic; o-m – oligo-mesotraphentic; m – mesotraphentic; me – meso-eutraphentic; e – eutraphentic

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Fig. 3. Surface plots for biological and environmental variables relationship analysis in the Kolsay lakes

Table 4.	Assessment of	of the ecological	state of the	Kolsay lak	kes in terms	of hydrophysic	cal and
		hydrochemi	ical paramet	ers, Augus	st 2015		

Variable	Lo	wer	Mic	ldle	Upj	ber	Kolsay		
	Ko	lsay	Kolsay		Kol	say	Sary-Bulak		
	Class	Rank	Class	Rank	Class	Rank	Class	Rank	
Secchi, m	Ι	1		1		1		1	
BOD, mgO/ dm ³	I	1	1	1	1	1	11	2	
$N-NO_2$, mg/ dm ³	III	4		4		4	11	3	
$N-NO_3$, mg/ dm ³	II	2	II	2	II	2	I	1	
N-NH ₄ , mg/dm ³	I I	1	II	3	11	3	11	3	
$P-PO_4$, mg/ dm ³	I I	1	1	1	1	1	I	1	
Point total	9	10	10	12	10	12	8	10	

3.4 Assessment of the Ecological State of the Lakes by Phytoplankton

Diatoms dominated in the phytoplankton communities in the three lower lakes, and green algae prevailed in the Sary-Bulak Lake (Fig. 4).

Most of the phytoplankton species from the general taxonomic list belonged to alkaliphilic and alkalibiontes that prefer water with pH higher than 7.0, as well as to indifferents [3]. More than half of planktonic species indicators (56%) were represented by category indifferent to the water

flow and oxygen saturation or prefer standing water bodies (Table 3). In relation to the mineralization, oligohalobes were dominant, among which the main part accounted for the indifferents.

The following changes occurred in the composition of phytoplankton with increase in altitude: the proportion of green algae species that live mainly in the plankton, the indicators of moderate temperatures, preferring standing water bodies and waters poorly enriched with oxygen, and alkaliphilic, increased; and the proportion of salinity indicators, with prevalence of oligohalobes – indifferents, decreased (Fig. 4).

Of the total taxonomic list of algae, saprobic valence is known for 21 species. The communities of all studied lakes were dominated by species - indicators of moderate pollution (Class III of water quality) (Fig. 5). The composition of phytoplankton in the Lower and Middle Kolsay included an equal number of species that are typical for pure and polluted water. The latter were recorded in the Upper Kolsay. The species composition of phytoplankton became more homogeneous in relation to organic pollution in the high-altitude direction.

The dominant species make the maximum contribution to the assessment of the level pollution. Blue-green of organic algae Planktolyngbya limnetica and Geitlerinema amphibium (Lower Kolsay) are characteristic for clean and moderately polluted waters. Lindavia comta (Bacillariophyta) prefers waters with low levels of organic pollution (Middle, Upper Kolsay and Sary-Bulak), and Cyclotella meneghiniana is an indicator of organic pollution (Lower, Middle and Upper Kolsay). Green algae Monoraphidium obtusum, M. contortum, and Closteriopsis longissima (Sary-Bulak) are characteristic for clean and moderately contaminated water.

According to the average values of saprobity index (Table 2), water of the Lower Kolsay was assessed as moderately polluted (β -mezosaprobic zone), Class III of water quality, and the Middle, Upper Kolsay and Sary-Bulak – as pure (oligosaprobic zone), Class II. Similar results were obtained when only diatoms were used for bioindication. According to Watanabe classification, the number of diatoms species, characteristic of pure water (saproxenes and eurysaprobes), increased, and the number of

saprophiles decreased in the high-altitude direction (Fig. 5).

Proportion of the eutrophic water indicators increased with altitude, but indicators of trophic state as a system are only known for diatoms, so the Sary-Bulak Lake was not assessed (Fig. 5). Indication of nutrition type, is represented by diatoms only, showed that the proportion of strictly autotrophic species (ats) and species, enduring a low organic load (ate), decreased from upper to lower lakes and the proportion of facultative heterotrophic (hne) increased (Fig. 5). The WESI index varied from 0.50 to 0.67 in the three lakes, showing suppression of algal communities and only in the Upper Kolsay Lake it was equal 1.

Canonical correspondence analysis of the relationship between the environmental variables and dominant species distribution over the lakes communities (Fig. 6a), or over the taxonomic Divisions (Fig. 6b) carried out on the basis of Tables 1, 2 and 3, showed significant differences between the Sary-Bulak Lake, Lower Kolsay, and two intermediate, similar to each other, Middle and Upper Kolsay lakes. Fig. 6 shows that the environmental indicators stating the degree of influence on the algal communities divide into the following groups: trophic (arrows pointing down), affecting to a greater extend the phytoplankton of three lower lakes and stimulating the development of blue-green algae and euglenoids, and inorganic, associated with ground-water flow and affecting the community of the Sary-Bulak Lake.

According to the values of phytoplankton biomass (Table 2), the waters of the Lower, Middle and Upper Kolsay lakes were assessed as very clean (Class II, Rank 2), and in the Sary-Bulak as extremely pure (Class I, Rank 1) [6].

For further analysis, indicators for size structure of phytoplankton were drawn with W value statistics of Clarke, the value of average cell mass, and the value of the Shannon-Weaver index (bit/individual, and bit/mg).

Cumulative graphs of dominant species of planktonic algae showed changes in the size structure of communities in the high-altitude direction (Fig. 7). In Lower Kolsay, the biomass curve was above the abundance curve, i.e. Wstatistic of Clarke had a negative value. The W value became positive and increased in the direction from the Middle Kolsay to upstream lakes.

The increase in the average cell mass (Table 2) also reflects the changes in the structure complexity of the phytoplankton in the highaltitude direction. In the Lower Kolsay, numerically dominant small-size blue-green algae occupied higher ranks (i.e. the proportion of the first dominant in the total abundance was higher than that of the first dominant in the total biomass) than larger diatoms, dominating in biomass (Table 5). Larger species of algae occupied higher ranks in biomass than in abundance for the phytoplankton of the three upstream lakes, respectively.







Fig. 5. The ratio of the species number - indicators of organic pollution, trophy and nutrition type in phytoplankton of the Kolsay lakes, August 2015. Abbreviation of ecological groups as in Table 3

Table 5. The average cell mass and the ranks of the dominant species in the total abundance
and biomass in phytoplankton of the Kolsay lakes, August 2015

Lake	Average cell mass ×10 ⁻⁶ , mg	Rank/Abundance, %	Average cell mass×10 ⁻⁶ , mg	Rank/ Biovolume, %
Lower	0.009	1/36.3	6.000	1/35.4
Kolsay	0.045	2/33.3	6.134	2/27.2
	0.014	3/20.6	5.224	3/15.5
Middle	6.134	1/37.7	6.134	1/38.5
Kolsay	8.475	2/24.4	8.475	2/34.4
	6.000	3/20.0	6.000	3/20.0
Upper	6.134	1/27.2	6.134	1/38.6
Kolsay	0.524	2/24.3	6.000	2/29.3
	6.000	3/21.2	8.475	3/17.8
	8.475	4/9.1		
Kolsay	17.667	1/28.2	17.667	1/74.5
Sary-	0.212	1/28.2	8.475	2/18.4
Bulak	8.475	2/14.5		
	1.706	2/14.5		
	0.524	2/14.5		





(b)

Fig. 6. Canonical Correspondence Analysis of relationship between the variables of the Kolsay lakes environment and the dominant species (a), and Divisions (b) in phytoplankton. Abbreviation of species names as in Table 3

Numeric expression of mutual position of dominance curves is the ratio of the Shannon-Weaver index values, calculated in two versions – bit/mg and bit/individual. The arithmetic difference between the values of the two versions was called Δ -Shannon-Weaver. There was a negative relationship between the amount

of W-value and values of Δ -Shannon-Weaver (Fig. 8).

4. DISCUSSION

Hydrochemical variables of the Kolsay lakes in August 2015 are corresponded to the levels of

extremely pure and clean water reservoirs (I – II Class, Ranks 1–3), except for nitrites. According to the total points calculation (Table 3), the organic pollution decreased in the line "Middle Kolsay = Upper Kolsay > Lower Kolsay > Sary-Bulak. Phosphorus was not found in water of all lakes, which is characteristic of extremely pure water, according to hydrochemical classification [6]. Comparison with the data of previous years showed that in August 2002 the amount of phosphates in the water of the Lower and Middle Kolsay (Fig. 9) has reached the level of slightly polluted water. During the reported period of 2002, the total nitrogen content in the water of the Lower Kolsay Lake has increased from 0.143 to 0.716 mg dm⁻³, and in the Middle Kolsay, on the contrary, decreased from 1.286 to 0.574 mg dm⁻³ [21].



Fig. 7. The structure of species dominance in phytoplankton of the Kolsay lakes, August 2015



Fig. 8. Dynamics of the W-statistic (Clarke) and \triangle -Shannon-Weaver

Along with temperature, phosphorus and nitrogen are essential elements for controlling the development of planktonic algae [9,51-53]. In August 2015, in the absence of phosphorus in the waters of all lakes, phytoplankton biomass varied similarly to the dynamics of total nitrogen concentrations, with the highest values in the Middle and Upper Kolsay. It should be taken into consideration that the phosphate concentration at a given time is not an indicator of the productivity of the system, but only points out to the active phosphorus exhaustion by phytoplankton. That is, the low contents of mineral phosphorus may indicate that the system is either exhausted, or its metabolism is intense [54]. Obviously, this is also true for the Lower

Kolsay, where over the last decade there was a reduction of phosphates to analytical zero, and the concentration of total nitrogen increased almost twice, indicating the nitrogen/phosphorus system imbalance, with the latter being exhausted. The emergence of Charophyta algae in the coastal zone of the lake (Fig. 10) shows the intensification of in-water processes that were absent in 2002.

The use of biological indicators allowed capturing more subtle variations in the quality of the Kolsay lakes water. The ratio of species in phytoplankton communities of the Kolsay lakes as a whole matched the one for non-flowing alkaline waters. In terms of the ratio of indicator



Fig. 9. The interannual dynamics of biogenic elements in the Kolsay lakes water



Fig. 10. The coastal area of the Lower Kolsay Lake in 2006 (left) and 2015 (right). Photo by Udartseva E.R. and Krupa E.G

species and the ratio of functional groups of phytoplankton (type of nutrition), the level of organic pollution decreased in the high-altitude direction. According to the values of phytoplankton biomass, the water of the Lower, Middle and Upper Kolsay was assessed as very clean, and of the Sary-Bulak – as extremely clean.

In addition to the species composition, the size structure of communities has significant importance. In nutrient-rich conditions, the dominance of small-sized species (r-strategists) is a general principle for functioning of the biological systems [54]. The domination of small-sized species in planktonic [10-13,21,55,56] and benthic communities [15-17] enhances in the process of eutrophication of water bodies. Dimensional structure of diatoms is a sensitive indicator of even short-term changes in aquatic ecosystems [57].

The ABC-Method is an integral expression of community size structure. R. Warwick [14] showed that the biomass curves are above the abundance curves in the normal habitat of benthic organisms. With moderate stress, the curves are close to each other; with strong stress, the abundance curves are located above the biomass curves.

The values of W statistic of Clarke, showing the relative position of the curves of biomass and abundance, indicate a change in the structure of phytoplankton in the Kolsay lakes with reduction of stress in a high-altitude direction. The value of the indicator changed synchronically with the dynamics of the average mass of algal cell in community, and the values of Δ -Shannon-Weaver. Previously, different statistically significant negative relationships between the Shannon-Weaver index and the relative amount of average algal cell size [10], as well as the average mass of an individual in zooplankton communities [21] were identified. A relationship between the W statistic values and Δ -Shannon-Weaver is due to the fact that they are based on the same principles as the relative proportion of species in abundance and biomass in communities. Therefore, information about the size structure of communities can be obtained by different methods such as graphic (ABC-Method) that ranged species abundance, and calculated ones (Δ-Shannon-Weaver and average individual mass/cell volume).

The WESI index ranged from 0.50 to 0.67 in the three lakes, and only in the Upper Kolsay was

equal to 1. Its values indicated some toxic effects on photosynthesis of algae in the Lower, Middle Kolsay and the Sary-Bulak. Heavy metal concentration in the waters of the lakes was on generally low level. Their minimum concentration (except for chromium and cadmium) was observed in the Upper Kolsay. An additional toxic factor having effect on planktonic algae can be high concentrations of iron in the three lakes. Iron is an essential element for the development of planktonic algae [58], but in high concentration has a negative impact on aquatic biota [59]. These results help us to assess the lakes ecosystems state as under early stage of toxic and nutrients impact that need for future monitoring with some traditional and advances methods [1,43,60].

The dynamics of biological indicators are linked well with the distribution of the recreational load on the lakes. The Lower Kolsay Lake, that has an automobile road leading to it, is under the greatest load. Other lakes can only be reached through the pathway, by foot or on horseback, so they are visited by a fewer tourists. This anthropogenic pressure on the lakes is growing every year. According to the official data, the number of tourists visiting the Kolsay lakes increased more than twofold from 6822 to 16000 people in the period from 2007 to 2015.

The net fishing was allowed here before the establishment of the National Park on the lakes. As a result, the near-bottom water layers of the Lower Kolsay Lake accumulated a large number of fine-mesh fishing nets that clung to the trunks of fir trees, once grown on the bottom of the valley. Fish that is captured and dies in fishing nets is the source of secondary enrichment of the water by nutrients and accelerates the process of eutrophication of Lower Kolsay Lake. Enrichment of ecosystem by nutrients is primarily manifested in the coastal shallow and more warmed-up areas of the lake. The results of future monitoring can be used as a base for the construction the regional program for the studied lakes watershed management that can to include not only hydrological and biotic data analysis but also economic and nature conservation relations [1,60].

The ongoing climate change can be taken into account in addition to the human factor. The global warming primarily influences the hydrological regime of water ecosystems in arid conditions of Kazakhstan, as well as in other regions [61]. The level of the Kolsay lakes has fallen during recent decades: the Upper Kolsay by 2.5 - 3.0 m, the Lower Kolsay – by 0.4 - 0.5m. The level of Middle Kolsay Lake, that benefits from the Kolsay River and two more small rivers that originates from the western slopes, has remained almost unchanged. The hydrological regime has both indirect and direct effect on hydrocenosis and water quality through the changes of depth, nutrient concentrations, and ratio of the bottom area to water volume. Generally, the symptoms of eutrophication of aquatic ecosystems are more clearly manifested in periods of low water levels [62,63] that we have observed in the studied lakes. For the Kolsay lakes, it stays unclear what is the major cause of eutrophication - an increase of recreational load or the climate warming and the following decline of their levels. Obviously, these two factors act together and can be monitored in the Kolsay lakes placed in protected areas. That means that we can manage the water quality in these lakes only as a result of monitoring with traditional as well as modern remote sensing methods [1,60].

5. CONCLUSION

The complex evaluation of the ecological state of the Kolsay high mountain lakes is set forth. According to hydrochemical variables, the lakes are classified as clean, with a slightly increased level of organic pollution in the Lower and Middle Kolsay. The ratio of species-indicators in the phytoplankton communities was reflected freshwater non-flowing alkaline waters. Regarding the ratio of species-indicators. the distribution of the functional groups (nutrition type) and indicators of size structure of phytoplankton, the level of organic pollution decreased in the high-altitude direction. The relationship between the W statistic of Clarke and the Shannon-Weaver index is shown. According to the values of phytoplankton biomass, the water of the Lower, Middle and Upper Kolsay was assessed as very clean, and of the Sary-Bulak Lake as extremely clean. Dynamics of saprobiological and dimensional parameters of phytoplanktocoenosis are linked well with the distribution of the recreational load on the lake.

The change in biomass of planktonic algae was determined to a greater extent by the dynamics of nitrogen compounds together with the exhaustion of phosphorus. Analysis of hydrochemical and biological data is demonstrated that the Kolsay mountain lakes are

in the early stages of eutrophication. The eutrophication processes are most evident in the Lower Kolsay. The values of the WESI-index indicated some toxic effects on photosynthesis of algae in the Lower and Middle Kolsay and the Sary-Bulak with generally low level of heavy metals content in the water. This conclusion is confirmed by the change in the concentrations of heavy metals and iron in the water of lakes. The changes in ecosystems of the Kolsay mountain lakes can be caused by the growth of recreational load with climate change in the background.

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COMPETING INTERESTS

Authors have declared that no competing interests exist.

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