# **Review Article**

# Critical environmental factors for photosynthetic organisms of the Shardara Reservoir, Kazakhstan

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# Keywords:

Phytoplankton, Macrophytes, Environmental factors, Shardara Reservoir, South Kazakhstan

#### Abstract

In summer 2015, the distribution of structural indicators of phytoplankton and the macrophytes overgrowth in the Shardara Reservoir's water independent of external factors was studied. Phytoplankton was represented by 78 species; green algae dominated. The abundance of community was 544.0 mln. cells m<sup>-3</sup>, with biomass at 626.1 mg m<sup>-3</sup>. Macrophytes Potamogeton natans L. and Potamogeton malajanus L. massively developed on the eastern shallow parts and in bays of the southwestern part of the reservoir. Our research showed that the biotopes inhabited by macrophytes were generally characterized by relatively higher concentrations of nitrite, phosphate, and zinc. The structure of phytoplankton was dependent on many factors, among which the most important were the water temperature and heavy metals. The warm-water status of the Shardara Reservoir was reflected in the dominance of green algae and dinoflagellates. With relatively high concentrations of heavy metals in the ecosystem, their impact on phytoplankton was neutralized by the complex nature of pollution, which included the increased number of organic substances. The impact of toxicants was not traced when analyzing the diversity and abundance of phytoplankton. The prevalence of facultative heterotrophs among the diatoms served as an indirect indication of the presence of toxic substances in the ecosystem. The dimensional structure of phytoplankton changed under the influence of cadmium in size reduction, and that may be the adaptive restructuring of the community in response to the toxic stress. Therefore, we may emphasize the essential indicative importance of size parameters of communities, including the Clarke's W-statistics and  $\Delta$ -Shannon-Weaver.

#### 1. Introduction

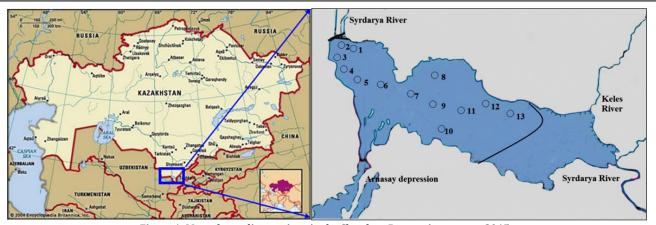
The Shardara Reservoir is located in the southernmost region of Kazakhstan, near the border with Uzbekistan. The region is characterized by hot climate, with maximum summer temperatures up to 49°C. Precipitation here averages 429 mm. Precipitation is the lowest in August, with an average of 2 mm. Most precipitation falls in March, with an average of 75 mm [1]. The reservoir was constructed in 1965 for irrigation purposes by damming the Syr Darya River. The river flows through four countries – Uzbekistan, Kyrgyzstan, Tajikistan, and Kazakhstan. During the summer, the water from the reservoir is used for irrigation of agricultural land along the southwestern and part of the northeastern coasts. There are also irrigated lands along the Syr Darya riverbed downstream of the dam. Water abstraction for irrigation leads to the decline in level and reduction of the reservoir area from spring to autumn about 3 times. The reservoir water level increases in autumn and winter.

Along with irrigation, the Shardara Reservoir serves as one of the largest fisheries of Kazakhstan. The cross-border nature of the Syr Darya River, agricultural development of the watershed region, its regional and climatic characteristics, as well as the reservoir being located in the metallogenic province [2], in combination with its unstable hydrological regime, are all responsible for a variety pollutants entering the water body and for the variability of their concentrations in time and space. The complex of natural and anthropogenic factors affects the entire ecosystem of the Shardara Reservoir. The most important of these are the nutrients and pesticides entering the reservoir from irrigation fields with return water and from the catchment area, as well as heavy metals that are part of some fertilizers washed from the catchment area. One of the sources of pollution is a trans-boundary runoff from the territory of the neighboring states.

Phytoplankton and macrophytes as primary producers determine the development of the upper links of the food chain in a water body and their conditions of survival. Information on phytoplankton and macrophytes of the Shardara Reservoir is absent in the literature. The purpose of this paper is to analyze the spatial distribution of planktonic algae and macrophytes in the waters of the Shardara Reservoir in accordance with environmental factors.

#### 2. Materials and methods

Research on water salinity and chemical composition was carried out in June 2015 in 3 stations, and research of the content of biogenic elements, heavy metals, and organo-chlorine pesticides in the water was carried out by means of a grid of 13 stations (Fig. 1). The measurements of temperature and pH of the surface water layers were taken in the field trip using a Hanna HI 98129 instrument. Water clarity was measured with Secchi disk. At each station, we visually assessed the degree of macrophytes' overgrowth on the water surface.



**Figure 1: Map of sampling stations in the Shardara Reservoir, summer 2015.** 1-13 are sampling stations. Black line outlines the water level of the reservoir during the sampling period.

One liter water samples to determine the salinity and ionic composition, the content of biogenic compounds, heavy metals, organochlorine pesticides, and the samples of phytoplankton were collected from the surface layer. Samples for the determination of nutrients were fixed with chloroform, for determination of heavy metals – with concentrated nitric acid.

Conventional methods of chemical analysis of water were used [3,4]. Water samples were analyzed in three-four replications. The error of estimate for major ions in the water was 0.5-5%, depending on the analyte.

Heavy metal measuring was performed by mass spectrometry with inductively coupled plasma using Agilent 7500 A, manufactured by Agilent Technologies, USA (National Standard RK ISO 17294-2-2006). The device allows for the detection of the various chemical elements in complex matrices, including those in the sea and grey water, and in the biological objects in micro-trace quantities. Abundance Sensitivity of Agilent 7500 A: Low Mass <5  $10^{-7}$ , High Mass <1  $10^{-7}$ . Data on the content of biogenic elements, heavy metals, and pesticides were compared to the maximum permissible concentrations of substances established for fisheries (MPC) [5, 6]. Organochlorine pesticides were measured by gasliquid and high-performance liquid chromatography [7-9]. Test-sensitivity is  $10^{-5}$  mcg. The detection threshold is 0.002 mg dm<sup>-3</sup> of the sample.

For the processing of phytoplankton samples, the settling method was used [10]. Species identification of planktonic algae was performed using determinants for relevant divisions [11-16]. Shannon-Weaver index was calculated on a Primer 5 program with the logarithm with base 2 in two versions: for the species abundance (bit individual<sup>-1</sup>), and for the species biomass (bit mg<sup>-1</sup>) [17]. Curves on domination were calculated using the same program. W-statistic of Clarke was calculated automatically [18]. Its value indicates the position of the curve biomass in respect to the curve of abundance. A positive W value indicates that the

biomass curve is higher than the 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.

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 Barinova *et al.* [19], 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 responded predictably to environmental conditions were used as bio-indicators for particular variables of aquatic ecosystems, the dynamics of which are related to environmental changes. The statistical methods used were those recommended by Heywood [20] for the development of taxonomic studies, namely, the CANOCO program [21] for canonical correspondence analysis (CCA) [22, 23], the GRAPHS program [24] for Principal Component Analysis (PCA), and comparative floristics. Surface plots for biological and environmental variables relationship analysis were constructed using the Statistics 12 program.

#### 3. Results

# 3.1. Hydrophysical, hydrochemical and toxicological characteristics of the reservoir

The reservoir is elongated from northwest to southwest (Figure 1). At the maximum filling, it has a water surface area of 783 km<sup>2</sup>. The right bank is more leveled, occasionally steep, composed of loose sand clay and clay loam, with steep underwater ledges (Figure 2). The left bank is flatter, dissected with bays and coves. The maximum depth does not exceed 20 m. Gray silt with sand and clay dominated the area, with small amounts of small-scale detritus in the upper parts of the reservoir. Contamination of soil by hydrogen sulfide was not found. Transparency varies across the waters from 0.50 to 3.20 m.



Figure 2: Shardara near the orographic right bank, summer 2015.

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The reservoir has warm-water, with an average water temperature during the study period exceeding 27°C (Table 1). The water is alkaline, with a slight variability of pH values across the water area. As follows from the ratio of ions and their total content, the water is classified as brackish, hard, as a sulfate class in the sodium group [25]. During the research, the content of biogenic elements was low. The highest concentrations of ammonium were recorded in the middle of the reservoir, and of nitrites – in the eastern shallow part near the right bank. In contrast, elevated concentrations of nitrates characterized the northwestern part of the reservoir, the Arnasay Bay area, as well as the area along the northeastern coast. The total content of nitrogen compounds reached maximum values in the northeastern shallow part of the water that is under the influence of the Syr Darya River runoff. The distribution of mineral phosphorus in the water area was relatively even, similarly maximizing its content in the northeastern shallow part of the reservoir.

Indicator	Units	Mean value	Indicator	Units	Mean value
Temp	°C	27.5±0.5	TDS	mg dm-3	1054.7±16.8
Depth	m	7.85±1.15	NH <sub>4</sub>	mg dm-3	0.115±0.020
Secchi	m	1.47±0.24	NO <sub>3</sub>	mg dm-3	5.45±0.13
Macrophyte	%	31.2±10.4	NO <sub>2</sub>	mg dm-3	0.09±0.03
рН		8.54±0.01	P-PO <sub>4</sub>	mg dm-3	0.040±0.012
Са	mg dm-3	107.9±1.9	Porg	mg dm-3	0.161±0.004
Mg	mg dm-3	60.0±0.4	Zn	mg dm-3	0.121±0.040
Na+K	mg dm-3	120.3±2.7	Cu	mg dm-3	0.040±0.006
HCO <sub>3</sub>	mg dm-3	177.0±1.8	Cd	mg dm-3	0.0032±0.0018
SO <sub>4</sub>	mg dm-3	494.2±11.1	Pb	mg dm-3	0.034±0.014
Cl	mg dm-3	78.6±0.6	НССН	mkg dm-3	0.0041±0.0008

Table 1: Hydrophysical, hydrochemical, and toxicological characteristics of the Shardara Reservoir, summer 2105.

The content of heavy metals was at an increased level (Table 1). The concentration of copper in the water exceeded the maximum permissible norms 35-65 times, of zinc – 4.3-23.6 times, of cadmium – 3 times [5, 6]. Of the organo-chlorine pesticides, HCH (hexachlorocyclohexane) was present throughout the water, with the highest concentrations along the southwestern shore of the reservoir and in the zone of the river flow. The highest zinc content was found in the shallow eastern third of the water area, the highest copper content – in the deep northwestern part, including the area near the dam along the western shore and the Arnasay Bay.

## 3.2. Macrophytes

Two types of macrophyte communities were represented with a domination of pondweed – by floating *Potamogeton natans* L., and Malayan *Potamogeton malajanus* L. During summer 2015 macrophytes did not have much overgrowth in the reservoir. Macrophytes developed in the eastern shallow part of the water and in a form of narrow stripes in the shallow waters near the northeastern coast and the bays of the southwestern part (Figure 3). The blocks of floating pondweed, *Potamogeton natans* L. (Figure 4), were recorded in the southwestern part of the reservoir. The coastal waters of the eastern part were marked by a broad band of another species of the same genus, *Potamogeton malajanus*.

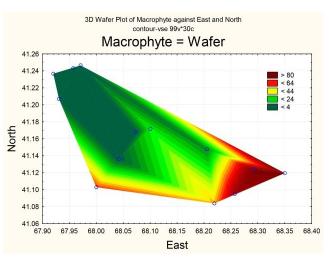


Figure 3: The spatial distribution of macrophytes in the waters of the Shardara Reservoir, summer 2015



Figure 4: Macrophytes of the Shardara Reservoir, summer 2015. Potamogeton natans (a), Potamogeton malajanus (b).

*Potamogeton natans* inhabits standing or slow-flowing water. *Potamogeton malajanus* prefers alkaline water with high temperature and of sufficient hardness.

# 3.3. Phytoplankton

Phytoplankton was represented by 78 species, including 42 species of greens, 16 of diatoms, 8 of blue-green, 7 of dinophytes, 3 of ochrophytes, and 2 of charophytes (Table 2). Diatoms *Fragilaria crotonensis*, *F. capucina*, *F. acus*, *Ulnaria ulna*, greens *Monoraphidium* 

contortum, M. griffithii, M. arcuatum, Monoraphidium minutum, Scenedesmus communis, S. obliquus, Myrmecia irregularis, Oocystis solitaria, O. lacustris, Pseudodidymo cystis planctonica, Planctonema lauterbornii, Tetradesmus lagerheimii, Tetraëdron caudatum, chrysophyceae algae Dinobryon elegans, cyanobacteria Merismopedia minima, dinophytes Peridiniopsis penardii, P. polonica, P. quadridens were found in the large part of water area.

Table 2: The species compos	sition and ecological character	istics of plankton algae in the Sh	ardara Reservoir, summer 2015

Species      Code      Intr. cell      mg m <sup>3</sup> , mg m <sup>3</sup> , mg m <sup>3</sup> , end      Hab      T      Oxy      Sal      D      Tro      Aut-H        Bacillariophyta	et pH ind alf alf alf alf alf alf ind alf ind alf ind alf alf
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Fragilaria crotonensis Kitton      FraCro      9.8      3.7      P      -      st-str      I      es      m      ate        Gomphonema parvulum (Kützing)      Kützing      GomPar      0.2      0.1      B      temp      str      i      es      o-m      hne        Lindavia contra (Kützing)      Nakov, Gullory,      LinCom      1.7      3.4      P      -      st      i      sx      o-m      hne        Melosira varians C. Agardh      MelVar      0.7      1.1      P-B      temp      st-str      hl      es      me      hne        Proschkinia longirostris (Hustedt)      D.G.Mann      ProLon      1.8      2.0      B      -      -      hl      -	alf ind alf alf alf - - ind
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Coenocystis planctonica Korshikov  CoePla  3.0  0.8  P  -  i  -  -    Coenolamellus botryoideus Proshkina-  CoeBot  2.2  0.7  P  -  h  -  -	-
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Crucigenia quadrata Morren CruQua 4.4 0.8 P-B - st-str i	acf
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Lagerheimia marssonii Lemmermann LagMar 0.4 0.1	-
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Myrmecia irregularis (J.B.Petersen) Ettl & MyrIrr 3.7 15.5	
Oocystis borgei J.W.Snow      OocBor      3.0      0.7      P-B      -      st-str      i      -	-

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<i>Oocystis lacustris</i> Chodat	OocLac	3.9	9.2	P-B	-	st-str	hl	-	-	-	
Oocystis solitaria Wittrock	OocSol	5.9	17.4	Р	-	st	i	-	-	-	ind
Pediastrum boryanum (Turpin) Meneghini	PedBor	5.9	5.4	P-B	-	-	i	-	-	-	
Pediastrum duplex Meyen	PedDup	1.5	1.3	Р	-	st-str	i	-	-	-	ind
Planctonema lauterbornii Schmidle	PlaLau	49.1	9.2	-	-	-	-	-	-	-	-
Pseudodidymocystis planctonica (Korshikov) E. Hegewald & Deason	PsePla	5.5	0.7	-	-	-	-	-	-	-	-
Scenedesmus apiculatus (West & G.S.West) Chodat	SceApi	0.7	0.6	Р	-	st-str	-	-	-	-	-
Scenedesmus obliquus (Turpin) Kützing	SceObl	19.3	20.1	P-B	-	st-str	i	-	-	-	ind
Schroederia setigera (Schröder) Lemmermann	SchSet	0.2	0.3	P	-	st-str	i	-	-	-	-
Stauridium tetras (Ehrenberg) E.Hegewald	StaTet	1.5	2.6	P-B	-	st-str	i	-	-	_	ind
Tetradesmus lagerheimii M.J.Wynne & Guiry	TetLag	10.0	3.9	P-B	-	st-str	i	-	-	-	ind
Tetradesmus obliquus (Turpin) M.J.Wynne	TetObl	3.7	2.4	P-B.S	-	st-su	i	-	-	-	-
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Ulnaria ulna (Nitzsch) P.Compère	UlnUln	2	2.3	-	-	-	-	-	-		-
Cyanobacteria	UIIUIII	2	2.5	-	-	-	-	-	-	-	
Aphanocapsa delicatissima West & G.S.West	AphDel	56.5	0.3	P-B	-		i	-	m	-	-
	Apridei	50.5	0.5	P-D	-	-	1	-	m	-	-
Aphanocapsa incerta (Lemmermann) G.Cronberg & Komarek	AphInc	69.4	0.3	P-B	-	-	i	-	me	-	-
Merismopedia minima G.Beck	MerMin	88.9	0.1	B,S	-	aer	-	-	0	-	-
Merismopedia punctata Meyen	MerPun	7.4	0.1	P-B	-	-	i	-	me	-	in
Microcystis aeruginosa (Kützing) Kützing	MicAer	13.1	0.9	Р	-	-	hl	-	e	-	-
Oscillatoria planctonica Woloszynska	OscPla	5.2	0.3	Р	-	-	i	-	me	-	-
<i>Planktolyngbya limnetica</i> (Lemmermann) Komarkova-Legnerova & Cronberg	PlaLim	23.7	0.2	P-B,S	-	st-str	hl	-	e	-	-
Trichodesmium lacustre Klebahn	TriLac	2.0	7.1	Р	-	st	-	-	-	-	-
Miozoa (Dinophyta)											
Ceratium hirundinella (O.F.Müller) Dujardin	CerHir	0.9	152.6	Р	-	st-str	i	-	-	-	-
Gonyaulax apiculata Entz	GonApi	0.4	8.1	-	-	-	-	-	-	-	-
<i>Gymnodinium variabile</i> Herdman	GymVar	1.7	3.1	P-B	-	-	mh	-	-	-	-
<i>Peridiniopsis penardii</i> (Lemmermann) Bourrelly	PerPen	3.5	34.5	Р	-	-	hl	-	-	-	-
<i>Peridiniopsis polonica</i> (Woloszynska) Bourrelly	PerPol	2.4	90.8	Р	-	st	i	-	-	-	-
Peridiniopsis quadridens (Stein) Bourrelly	PerQua	3.0	49.1	Р	-	-	-	-	-	-	-
Prorocentrum lima (Ehrenberg) F.Stein	ProLim	0.4	1.8	-	-	-	-	-	-	-	-
Ochrophyta (Chrysophyta)											
Dinobryon elegans Reverdin	DinEle	13.5	18.9	-	-	-	-	-	-	-	-
Dinobryon sertularia Ehrenberg	DinSer	1.7	1.5	Р	-	-	i	-	-	-	-
Mallomonas sp.	Mallom	0.6	8.5	-	-	-	-	-	-	-	-
Achnanthes gibberula Grunow	AchGib	1.3	6.9	В	eterm	st-str	mh	SX	o-m	-	in
Note: Temperature preferences (T): temp – te Dxygenation and water moving (Oxy): st – st [26]: alf – alkaliphiles, ind – indifferents; acf – - mesohalobes. Organic pollution indicators a oligotraphentic; o-m – oligo-mesotraphentic; (Lt) [20], otc. mitagan automphin two	anding water, acidophiles; a according to V m – mesotrap	str – streaming wa lb – alkalibiontes. H Vatanabe <i>et al</i> . [28 hentic; me – meso-	ter, st-str – low alobity degree ] (D): sx – sapr eutraphentic; e	v streaming v according Hu oxenes, es – – eutraphen	water, aer ustedt [27] eurysapro tic; o-e – o	– aerophil (Sal): i – o bes, sp – ligo-eutra	es. Acio oligohal saprop phentic	dity (p obes-ir hyles. ' . Nitrog	H) degre different Trophic s gen uptal	e according F , hl – halophi state (Tro) [2 se metabolism	Husteo iles, m 29]: o n (Au
Het) [29]: ats – nitrogen-autotrophic taxa, t concentrations of organically bound nitrogen; nce – nitrogen-heterotrophic taxa, needing ele	hne – facultat	tively nitrogen-hete	rotrophic taxa,	needing per	iodically el	evated co	ncentrat	ions o	f organica		

The average abundance of phytoplankton was at a relatively high level, with moderate value of biomass (Table 2). Cyanobacteria and green algae shaped the basis of abundance. Dominant complex included bluegreen algae *Merismopedia minima*, *Aphanocapsa incerta*, *Aphanocapsa delicatissima*, *Planktolyngbya limnetica* and green *Planctonema lauterbornii*. The percentage of these species accounted for 50% of the total abundance. Dinophytes dominated in terms of biomass, *Ceratium hirundinella*, *Peridiniopsis polonica*, *P. quadridens*, *P. penardii* together contributing 50%.

Table 2: Quantitative indicators of phytoplankton in the Shardara
Reservoir, summer 2015

Division	Abundance, mln. cells m <sup>-</sup> <sup>3</sup>	Biomass, mg m <sup>-3</sup>				
Bacillariophyta	37.4±5.4	90.1±19.8				
Charophyta	1.8±0.8	10.5±4.4				
Chlorophyta	210.2±26.6	153.9±20.1				
Cyanobacteria	266.5±168.8	2.8±1.4				
Miozoa (Dinophyta)	12.3±2.5	340.0±88.6				
Ochrophyta (Chrysophyta)	15.7±3.5	28.9±5.8				
Total	544.0±174.5	626.1±76.6				

The values of Shannon-Weaver index ranged from 2.90 to 4.69 bit individual<sup>-1</sup> and from 2.44 to 4.00 bit mg<sup>-1</sup> and showed, on average, an increased level of diversity of phytoplankton in the Shardara Reservoir (Table 4). The average weight of the algal cell characterized the small-sized composition of phytoplankton. The lowest value of the latter indicator was observed along the southwestern shore of the reservoir. The values of W-Clarke were negative, indicating that the abundance curve was above the biomass curve.

Table 4: Structural indicators of phytoplankton in the Shardara

Reservoir, summer 2015				
Indicator	Value			
N-Shannon	3.80±0.19			
B-Shannon	3.41±0.20			
∆-Shannon	-0.38±0.21			
W-Clarke	-0.095±0.040			
average cell mass, mg10 <sup>-6</sup>	1.66±0.27			

The correlation analysis showed that the total diversity of phytoplankton (with a major contribution being made by charophytes and green algae) grew on shallow warmed water areas overgrown with macrophytes and with a high concentration of nitrates and phosphates (Table 5).

The positive link between the diversity of charophytes algae and zinc was revealed. These shallow and well-warmed parts of the reservoir showed an increased abundance and biomass of charophytes and green algae. The abundance of blue-green algae was decreasing in the gradient of lead concentrations, and the total biomass of the species of this Division has been higher in the warmer water areas. Dinophyte abundance was increasing within the gradient of ammonium concentration, and the species biomass of this division was increasing in areas with higher transparency and high concentration of copper and cadmium while decreasing under the influence of zinc. Ochrophytes, mainly chrysophytes, abundance was higher in the deep part of the reservoir, poorly overgrown with macrophytes, with high water transparency and increased concentrations of HCCH. The increase of the average weight/volume of algal cell was occurred mostly due to diatoms and dinophytes, and in turn helped to increase the total biomass of phytoplankton. The values of the Shannon index were calculated according to the species abundance and biomass of phytoplankton. They decreased with a rise of water transparency and copper and cadmium content in the water, and increased within the gradient of concentrations of zinc, nitrites, and phosphates. The growth of charophytes and green algae biomass served as an internal factor contributing to the increase in the Shannon index values. Strong positive correlation was found between the values of  $\Delta$ -Shannon and W-Clarke. The values of both indices declined under the influence of cadmium and with an increase of the total biomass of phytoplankton, which in turn was determined by the abundance of dinoflagellates. Saprobity index SI values (that characterize the general level of organic pollution of the water body according to phytoplankton) increased in shallow, well-warmed waters with low-water transparency, overgrown with macrophytes.

Table 5: The coefficients of Spearman Rank Order Correlations between the abiotic characteristics and the phytoplankton structure a	nd
indicators group in the Shardara Reservoir, summer 2015. Negative correlations are marked in bold. Abbreviations as in Table 2.	

Paired indices	Spearman Rank Order Correlations	Paired indices	Spearman Rank Order Correlations	
Charophyta species – Temperature	0.861	B Chlorophyta – Nitrite	0.745	
Charophyta species – Depth	-0.894	B Chlorophyta – Phosphate	0.762	
Charophyta species – Secchi	-0.898	B Total – Cd	0.690	
Charophyta species – Macrophytes	0.964	Δ-Shannon – Cd	-0.979	
Charophyta species – Zn	0.820	Δ-Shannon – B-Miozoa (Dinophyta)	-0.700	
Charophyta species – Nitrite	0.823	Δ-Shannon – N-Miozoa (Dinophyta)	-0.667	
Charophyta species – Phosphate	0.786	Δ-Shannon – B-Total	-0.733	
Chlorophyta species – Depth	-0.673	Δ-Shannon – W-Clarke	0.917	
Chlorophyta species – Phosphate	0.694	Avg_cell_size – B-Bacillariophyta	0.683	
Total species – Macrophytes	0.708	Avg cell size – B-Total	0.833	
Total species – Nitrite	0.688	Avg_cell_size – B-Miozoa (Dinophyta)	0.733	
Total species – Phosphate	0.770	W-Clarke – Cd	-0.851	
Index S – Temperature	0.753	W-Clarke – B-Total	-0.667	
Index S – Depth	-0.817	P - Pb	0.735	
Index S – Secchi	-0.837	P-B – Phosphate	0.672	
Index S – Macrophytes	0.743	B – Temperature	0.781	
N-Charophyta – Temperature	0.861	B – Secchi	-0.773	
N-Charophyta – Depth	-0.894	temp – Zn	0.671	
N-Charophyta – Secchi	-0.898	temp – Ammonia	-0.748	
N-Charophyta – Macrophytes	0.964	temp – Nitrite	0.748	
N-Charophyta – Zn	0.820	temp – Polyphosphate	0.674	
N-Charophyta – Zh	0.823	i – Nitrite	0.691	
	0.825	i – Phosphate	0.891	
N-Charophyta – Phosphate			-0.737	
N-Cyanobacteria – Pb	-0.695	hl – pH		
N-Miozoa(Dinophyta) – Ammonia	0.720	hl – Ammonia	0.677	
N-Ochrophyta (Chrysophyta) – Depth	0.849	hl – Nitrate	0.802	
N-Ochrophyta (Chrysophyta) – Secchi	0.717	sx – Zn	0.677	
N-Ochrophyta (Chrysophyta) – Macrophytae	-0.823	sx – Cu	-0.757	
N-Ochrophyta (Chrysophyta) – HCCH	0,751	sx – Cd	-0.702	
B-Shannon – Secchi	-0.668	sp – HCCH	-0.728	
B-Shannon – Zn	0.895	m – Temperature	0.786	
B-Shannon – Cu	-0.736	m – Depth	-0.820	
B-Shannon – Cd	-0.701	m – Secchi	-0.842	
B-Shannon – Nitrite	0.807	m – Macrophytes	0.903	
B-Shannon – Phosphate	0.824	m – Zn	0.857	
B-Shannon – B-Charophyta	0.760	m – Ammonia	-0.674	
B-Shannon – B-Chlorophyta	0.778	m – Nitrite	0.861	
B-Charophyta – Temperature	0.732	m – Phosphate	0.861	
B-Charophyta – Depth	-0.771	e – Pb	-0.749	
B-Charophyta – Secchi	-0.788	ind – Zn	0.682	
B-Charophyta – Macrophytes	0.864	ind – Nitrite	0.729	
B-Charophyta – Zn	0.853	ind – Phosphate	0.817	
B-Charophyta – Nitrite	0.857	alf – Secchi	-0.732	
B-Charophyta – Phosphate	0.884	st – Depth	-0.798	
B-Cyanobacteria – Temperature	0.739	st – Macrophytes	0.682	
B-Miozoa(Dinophyta) – Secchi	0.728	st – HCCH	-0.788	
B-Miozoa(Dinophyta) – Zn	-0.683	st-str – Phosphate	0.710	
B-Miozoa(Dinophyta) – Cu	0.683	ats – Zn	0.725	
B-Miozoa(Dinophyta) – Cd	0.690	ats – Ammonia	-0.728	
B-Chlorophyta – Temperature	0.728	ats – Nitrite	0.728	
B-Chlorophyta – Depth	-0.933	ats – Phosphate	0.728	
B-Chlorophyta – Secchi	-0.879	ate – Secchi	0.681	
B-Chlorophyta – Macrophytae	0.853	ate – Macrophytae	-0.675	
B-Chlorophyta – Zn	0.767	ate – HCCH	0.810	

Among the type of habitat's association indicators, planktonic and plankton-benthic species were stimulated by increased water temperature and the presence of phosphates, whereas benthic species preferred more transparent water (Table 5).

It is notable that in places where the water had a substantial amount of lead, planktonic species gained the advantage. Temperature indicators were present only among the diatoms. Most of them preferred moderate water temperature. The indicators of organic pollution by Watanabe have been identified as among the diatoms only. Therefore, statistically significant Spearman correlation coefficient values showed that, in general, the development of diatoms was positively affected by increased concentrations of nitrates, polyphosphates, and zinc ions, whereas copper, cadmium, ammonium and HCCH inhibited their development.

Among the indicators of water mass mobility and oxygen enrichment, moderately oxygenated water was preferred by predominant species. However, their development was associated only with the phosphates in water. The number of species indicators of poorly oxygenated waters was positively correlated with the presence of macrophytes. HCCH affected their development negatively, more so with the increase of the depth.

Indicators of proton concentrations were mainly represented by the pH-indifferents, for which there was a positive correlation with zinc, nitrites, and phosphates. Indicators of mildly alkaline waters favored the areas with low-water transparency, which indicates the acidification of those habitats where the transparency was reduced.

Among water salinity indicators, the indifferents prevailed, with fewer quantities of halophilic species. Algae, indifferent to the salinity of water, showed a positive association with the presence of phosphates and nitrates in the water. Species that prefer high-chloride content positively responded to elevated concentrations of nitrate and ammonium, and negatively – on the increase of pH in the water.

An important indicator of the reservoir's trophicity was represented by indicators from meso- and eutrophic groups. Development of mesotrophes was maintained by elevated temperature and by the presence of macrophytes, nitrites, phosphates, and zinc in water. Indicators of the mesotrophic state preferred shallow water areas with a low content of ammonium. Indicators of eutrophic waters reacted negatively to the increase of lead concentration in the water.

The feed type of algae indicates the predominance of either a photosynthetic or facultative-heterotrophic way of building proteins. Among the feed type indicators, the autotrophs dominated; they could sustain substantial concentrations of nitrates in the water. Strictly autotrophic types were supported by nitrites, phosphates, and zinc; however, ammonium inhibited their development. More nitrate-tolerant autotrophs positively reacted to the increase in the water transparency and the presence of HCCH, but macrophytes competed with them for the nutrient resource. It is noteworthy that, for the indicators of the mixed feed type, there has been no substantial correlation with habitat indicators identified.

Canonical correspondence analysis showed that according to the degree of influence on the dominant species of algae (comprising more than 50% of the phytoplankton population), external factors were divided into two groups (Figure 5).

Zinc, the amount of which increased in the shallow warm waters overgrown with macrophytes, with low transparency and with an increased amount of phosphates and nitrates, had influence on the total quantity of planktonic algae. The most resistant to this group of factors was the community on station 10 (southwestern coastal zone, see Fig. 1) with blue-green *Aphanocapsa incerta* dominating in abundance (Figure 5, blue circle).

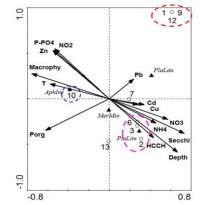


Figure 5: Canonical correspondence analysis of relationship between the variables of the Shardara Reservoir environment and the dominant species in phytoplankton, more than 50% of total abundance. Abbreviation of species names as in Table 2.

The second group of factors was represented by cadmium and copper, whose concentrations increased in the deep parts of the water body with high transparency. This group of factors also includes nitrates, ammonia, and pesticides. Copper, cadmium, pesticides, and nitrates in combination with hydrophysical factors had the greatest impact on the blue-green algae *Planktolyngby alimnetica* and *Merismopedia minima* (Figure 5, the pink circle), that dominated at stations 2, 3, and 6 (area near the dam).

We should also mention the communities dominated by green algae *Planctonema lauterbornii* that were not abundant and developed at the stations 1, 9, and 12 in the relatively shallow, warmed water habitats and under influence of the similar values of the rest of environmental factors (Figure 5, the red circle).

CCA analysis of algae divisions was carried out on the basis of Tables 1 and 2. Environmental variables formed two clusters that affect the Division structure of phytoplankton (Figure 6). The first cluster included Zn, organic pollution (according to the index S), and the water temperature. This group of factors had a stimulating effect on the species Charophyta and Chlorophyta in communities on stations 9-13. Cd, Cu, Pb, and HCCH formed a second group of factors collectively affecting the species richness of diatoms and dinophytes, and to a lesser extent of blue-green algae at stations 1, 2, 3, 6, and 7. In particular, for charophytes, a combination of factors from the second group had a fatal impact, and they were absent in the community composition on these stations. It is noteworthy that the depth and transparency of water played a negative role in the formation of phytoplankton communities' diversity and their abundance.

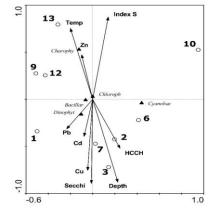


Figure 6: Canonical Correspondence Analysis triplot of relationships between the pollution variables of the Shardara Reservoir and phytoplankton Divisions. The bold numbers are the number of sampling stations as in Figure 1.

According to the CCA analysis, natural factors included water temperature, organic contamination expressed in index saprobity S, and Zn, while Cu, Cd and HCCH come from anthropogenic sources. It is noteworthy that the most typical stations are located in the northwestern part of the reservoir near the dam, where most of the pollutants are found in high concentrations. This is due to the movement of water masses in the direction from east-to-west and, as such contaminants as HCCH do not disintegrate in water, they move with the body of water. The impact of HCCH on the indicator species of phytoplankton in the Shardara Reservoir has been identified by means of CCA statistics.

## 4. Discussion

The Shardara Reservoir is located in the zone of intensive agriculture, and therefore its biota is exposed to the complex of natural and anthropogenic environmental factors. During the research, the water temperature reached, on average,  $25.5^{\circ}$ C, the depth was 7.85 m, water transparency – 1.47 m. The ammonium content had an average of 0.115 mg dm<sup>-3</sup>, nitrate – 5.54 mg dm<sup>-3</sup>, nitrite – 0.09 mg dm<sup>-3</sup>, phosphates – 0.040 mg dm<sup>-3</sup>, zinc – 0.121 mg dm<sup>-3</sup>, copper – 0.040 mg dm<sup>-3</sup>, cadmium – 0.0032 mg dm<sup>-3</sup>, lead – 0.034 mg dm<sup>-3</sup>, and HCCH isomers – 0.0041 mkg dm<sup>-3</sup>. All these factors formed two groups with regard to their impact on phytoplankton. Based on CCA analysis, it was concluded that copper, cadmium, lead, and HCCH isomers are coming from anthropogenic sources, and the temperature and the saturation of water by zinc ions are natural factors.

For the Shardara Reservoir, as one of the leading natural factors that determine the composition of the photosynthetic communities, represented by algae and macrophytes, can be the water temperature. In summer 2015, its value reached 38-39°C in the coastal zone. The warm-water status of the reservoir in the period of study was reflected by the dominance of green algae, both the number of species and abundance. Similar dominance of green algae was recorded for subtropical lakes Santragachi and Shibpukur [30, 31]. In large deep Alpine lakes such as Garda, Iseo, Como, Lugano, and Maggiore a high positive relationship between water temperature and Chlorophyta, Charophyta and Dinophyta algal Divisions was found [32]. We revealed similar relation between water temperature and the same algal Divisions that represented the majority of the Shardara Reservoir phytoplankton.

Among natural factors, the depth and transparency of water both played a negative role in the formation of diversity of phytoplankton communities and their abundance. This is likely to be caused not so much by the influence of hydrophysical conditions on phytoplankton, as by the other abiotic factors that correlated with them -the degree of overgrowing by macrophytes, the distribution of nutrients, heavy metals, and pesticides in the waters of the reservoir. With depths predominantly over 2.0 m, the pondweed developed massively in the eastern shallow part and in the shallow Arnasay Bay (Figure 1). It should also be noted that both of these areas are being influenced by additional pollutants coming from the Syr Darya and Keles rivers and by the discharge of drainage water, respectively. Evidently, that is why the biotopes overgrown by pondweed were characterized by higher concentrations of nitrites, phosphates, and zinc and by saprobic index values in comparison to the rest of the area. The increase in diversity and quantity of charophytes, total diversity of phytoplankton and abundance of green algae on shallow overgrown areas indicated that the complex of the said factors was favorable for these groups.

Along with the elevated values of saprobity index of phytoplankton, a higher trophic status of shallow water areas was evidenced by the environmental preferences of the dominant species of macrophytes. So, *Potamogeton natans*, prevailing in the shallow parts of reservoir, usually inhabits meso-, eutrophic, hypertrophic [33], non-flowing, and slightly alkaline waters, accumulating organic and mineral substances in the sediments [34].

150 cm with the optimum at 60-120 cm, on muddy, silt-sandy, and siltpeaty substrates. This species is resistant to fluctuations of water temperature and the density of its thickets depends on the depth. *Potamogeton natans* is able to develop in conditions of summer and autumn's low-water level; however, it serves as an indicator of the lack of summer water rise. It also forms rather dense thickets under the conditions of water level fluctuations. Increased anthropogenic eutrophication of the reservoir reduces the vitality and productivity of populations. The species are resistant to the reduced water transparency and is stimulated by a small influx of nitrogen fertilizers [34]. *Potamogeton natans* is resistant to organic pollution, it is an oligo-betamezosaprobiont with a saprobity index of 1.5 [35]. In other words, the massive development of this species is indicative of relatively lower trophic status of the reservoir water than according to the assessment of phytoplankton.

It grows in shallow water with a water depth between 5 and

The abundance of macrophytes correlated with the number of indicators of poorly oxygenated waters as well as indicators of mesotrophic species. This may signal the periodic fatal phenomena in the shallow overgrown parts of the reservoir. At the same time, macrophytes were competing with autotrophic algae for nutritional resources. The presence of such competition is also evidenced in the experiments of [36].

The second group of factors included copper, cadmium, lead, and HCCH isomers. According to the degree of toxicity, heavy metals form the following order for the phytoplankton Cd≈Cu>Zn>Pb [37, 38]. With high concentrations of copper, especially in the southwestern and northwestern deep areas, there was no negative impact on the diversity and quantitative indicators of phytoplankton communities revealed. In conjunction with cadmium, copper had a positive effect on the biomass of dinoflagellates and, to the species of this Division, on the total phytoplankton biomass.

This reaction of the Shardara Reservoir phytoplankton to heavy metals, including the most toxic ones – copper and cadmium, can be caused by several factors. One of these factors is the naturally elevated level of copper in water bodies of south and southeastern Kazakhstan [39] and the adequate adaptation of the regional flora populations to this metal. Many species of algae, certain diatoms in particular, are capable of enduring high copper concentration in the environment – up to 0.2-0.5 mg dm<sup>-3</sup> [40], which is substantially higher than the amounts of this metal in the Shardara Reservoir. According to the above-mentioned work, cadmium was less toxic and, at concentrations up to 0.2 mg/dm<sup>3</sup>, had no effect on diatoms. Relatively high resistance of plankton algae to heavy metals is conditioned by their ability to increase the production of intracellular polysaccharides in response to the pollution of the environment.

Studies of the effect of heavy metals on the diatoms have shown that, depending on the species, season, type of metal and other factors, the toxicity effect was different [41]. Diatoms *Achnanthes minutissima* and *Ulnaria ulna* preferred lower concentrations, and *Cyclotella meneghiniana*, on the other hand, favored high concentrations of cadmium in the environment. The direction of the link between *Ulnaria ulna* and copper depended on the season – in winter this relationship was negative, but it was positive in spring.

With Cd and Cu representing the highest danger for phytoplankton, it was shown [37] that the toxicity of these metals substantially declined in the presence of detergent components such as Nitrilotriacetic acid in the environment. This is of relevance to the Shardara Reservoir, where the detergent components come with a transboundary flow, as well as from the settlements located along the Keles riverbed and from the Saryagash resort. The contamination of reservoir water with detergents points to a ubiquitous presence of polyphosphates in the water. Thus, it may be another factor explaining the absence or small number of significant correlations between the diversity and a quantity indicators of phytoplankton and heavy metals.

One final important point is that metal toxicity declines when the trophic status of natural water increases [42]. The authors of the abovementioned work showed how the copper toxicity for periphyton diminished because of the higher concentrations of phosphorus in the environment, phosphorus serving as a key element accelerating the processes of eutrophication of the water body. Taking into account regional climatic and chemical indicators, the Shardara Reservoir is a mesotrophic water body with signs of increased trophic status in its shallow parts. The dominance of green algae in variety and quantity indicators, the small-sized composition of the community, which was reflected in values of the average, cell volume (Table 4), as well as the negative values of Clarke's W-statistics, all evidenced the elevated levels of organic pollution of the reservoir. Just as for other water bodies, the dominance of green algae [43] against the background of low values of dimensional structure [44-46] represents one of the signs of eutrophication of the Shardara Reservoir.

The values of the Shannon-Weaver index, calculated from the percentage of species in the total biomass, characterized a generally high level of diversity of phytoplankton in the reservoir, registered in other water bodies with high organic load [43]. There is a robust negative link between the Shannon-Weaver index (bit mg<sup>-1</sup>) and the average cell volume of algae [45, 47]. This negative relationship between the diversity index (bit mg<sup>-1</sup>) and the dimensional structure, which was also true for zooplankton was represented in our previous studies [48]. For phytoplankton in the Shardara Reservoir, the relationship between average cell volume and the Shannon index (bit mg<sup>-1</sup>) was negative; however, it had no statistical significance. This could also be caused by many factors acting multidirectionally on the biota of the reservoir.

One such factor that affects the size structure of the community is toxic pollution. We found a negative relationship between cadmium concentration and the averaged cell size in phytoplankton, and correlation between each other – Clarke's W-statistics and  $\Delta$ -Shannon-Weaver. We introduce the latter parameter as an arithmetic difference between the two versions of the Shannon-Weaver index [47]. Our results show that under the influence of cadmium, as one of the metals most toxic for living cells, there were internal structural adjustments within phytoplankton communities, aimed at strengthening the dominance of small-sized species. The experiments showed a decrease in the average cell mass (i.e., the increase in relative cell surface area) of the algae Coscinodiscus granii L.F. Gough exposed to cadmium and zinc [49]. That is, an increase in the cell surface, correlated with an increase of the membrane exchange, indicates the development of a protective mechanism in the phytoplankton communities in the Shardara Reservoir when exposed to toxic metals.

As a result, we found the complex nature of contamination that provides a protective function for the phytoplankton communities of the Shardara Reservoir. The impact of toxicants was not traced when analyzing the diversity and abundance of phytoplankton but was evident in the analysis of the feed type of algae. The presence of a toxic effect on the ecosystem of the reservoir was indicated by the prevalence of facultative heterotrophic species of algae in the water area (Table 2), however, with no statistically significant association with heavy metals (Table 5).

The most abundant macrophyte in the Shardara Reservoir – *Potamogeton natans* – grew in shallow water in its optimal conditions – on sandy and silty bottom, with relatively low inflow of organic pollution, and served as an indicator of the lack of the summer rise in water level, which is characteristic of the arid water bodies.

#### 4. Conclusion

Phytoplankton of the Shardara Reservoir was represented by 78 species, green algae dominated. The average abundance of community was 544.0 mln. cells m-3, with a biomass of 626.1 mg m-3. The index of Shannon-Weaver averaged out at 3.80 bit individual<sup>-1</sup> and 3.41 bit mg<sup>-1</sup>. Average cell mass/volume of algae was 1.66 10<sup>-6</sup> mg. Macrophytes Potamogeton natans L. and Potamogeton malajanus L. massively developed on the eastern shallow parts and in the bays of the southwestern part of the reservoir. Our research showed that the biotopes inhabited by macrophytes were generally characterized by relatively higher concentrations of nitrite, phosphate, and zinc. The structure of phytoplankton in the Shardara Reservoir depended on many factors, among which the most important were the water temperature and heavy metals. Temperature, along with salinity, determines the floral character of any water body. Warm-water status of the Shardara Reservoir is reflected in the dominance of green algae and dinoflagellates. With relatively high concentrations of heavy metals in the ecosystem, their impact on phytoplankton and macrophytes was neutralized by the complex nature of pollution, which includes the presence of organic substances, detergent ingredients and meso-eutrophic status of the reservoir. The impact of toxicants was not traced when analyzing the diversity and abundance of phytoplankton. The prevalence of facultative heterotrophs among the diatoms served as an indirect indication of the presence of toxic substances in the ecosystem. The dimensional structure of phytoplankton changed under the influence of cadmium towards cell size reduction, and that may be the adaptive restructuring of the community in response to the toxic stress. Therefore, we may emphasize the essential indicative importance of cell size parameters of communities, including the Clarke's W-statistics and  $\Delta$ -Shannon-Weaver. The most abundant macrophyte in the Shardara Reservoir Potamogeton natans grew in shallow water in its optimal conditions on sandy and silty bottom, with relatively low inflow of organic pollution, and can serve as an indicator of the lack of the summer water level rising, which is characteristic of the arid water bodies.

#### Acknowledgements

The work was carried out under project Nº 1846/ $\Gamma\Phi4$   $\Gamma.2015$ - $\Gamma2016$  for the Committee of Science, Ministry of Education and Science, Republic of Kazakhstan "Development of the methods for controlling the ecological state of water bodies in Kazakhstan" as well as partly supported by the Israeli Ministry of Absorption.

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