

## Functional and morphological groups in the phytoplankton of large reservoirs used for aquaculture in Bulgaria

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### Abstract

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Functional and morphological concepts for the classification of phytoplankton are increasingly used in environmental studies of freshwater ecosystems. The present study focuses mainly on the application of the three approaches (**FGs**, **MFGs**, **MBFGs**) for morpho-functional classifications used for detecting changes in phytoplankton communities in five Bulgarian large reservoirs that have long been used for aquaculture. Totally 113 taxa of planktonic algae were identified. The representatives of Cyanoprokaryota (**H1**, **M**; **5a**, **5b**, **5e**; **III**, **VII**), Chlorophyta (**J**; **3b**, **9b**; **IV**), Streptophyta (**P**, **N**; **8a**; **IV**) and Bacillariophyceae (**P**, **MP**, **D**; **6b**; **VI**) had the largest contribution to the abundance of algae in the studied reservoirs. Forty-two dominant species from eighteen functional groups (**FGs**), fifteen morpho-functional groups (**MFGs**) and seven morphologically based functional groups (**MBFGs**) were found. The functional groups with most species were: **H1**, **J**, **N**, **Lo**, **P** и **M**, most common among **MFGs** were: **5a**, **5b**, **5e**, **6b**, **8a** and **9b**, and among **MBFGs**: **III**, **IV**, **VI** and **VII**. The large gelatinous colonial (**M**; **5b**; **VII**) and filamentous (**H1**; **5a**, **5e**, **III**) cyanoprokaryotes, some of which are potentially toxic species indicative of eutrophication, were among the most common taxa in three of the studied reservoirs. According to the presented study, the three morpho-functional environmental concepts provide an important tool for understanding the seasonal changes in phytoplankton communities, as well as for assessing the environmental status of heavily modified freshwater reservoirs.

*Keywords:* phytoplankton, functional group, morpho-functional group, morphologically based functional groups, reservoirs

*Abbreviations:* **FGs** – functional group; **MFGs** – morpho-functional group; **MBFGs** – morphologically based functional groups

### Introduction

In recent years, when studying phytoplankton communities, some researchers have adopted an alternative scheme based on presenting the functional relationships between species. In freshwaters, the coexistence of species with similar environmental needs and similar requirements to the environment inhabited by them is called functional group (Reynolds et al., 2002). One of the first attempts to formulate a phytoplankton classification system based on its functional qualities was pro-

posed by Reynolds (1980). Nowadays, the functional group approach based on the physiological, morphological and environmental characteristics of the species proves to be more effective in analyzing the seasonal changes in phytoplankton biomass (Becker et al., 2010). Fourteen groups of phytoplankton were identified in the original study by Reynolds (1980).

The subsequent list of functional groups included 31 functional groups of phytoplankton (Reynolds et al., 2002), and additions and changes thereto were made by Padisak et al. (2009). Presently, phytoplankton functional groups are more than 40

(Reynolds, 2006; Padisak et al., 2009). The role of seasonal changes in physical processes and the selection of functional groups in environmental status assessment are also important (Becker et al., 2009). Both taxonomic and functional classification schemes show significant results, but the classification based on functional connectivity has exceptional capabilities (Kruk et al., 2010).

For the first time in Bulgaria the functional classification of phytoplankton was applied for monitoring of lakes and reservoirs for the period from 2011 to 2012 by Stoyneva et al. (2013). Results of the application of functional classification and similar conclusions were published by Belkinova et al. (2014). The perspective of their application was shown in the studies conducted by Stoyneva (2014) and Belkinova et al. (2014). Besides functional groups, there are two more environmental concepts that provoke special interest among researchers these days: morpho-functional groups (MFGS, Salmaso and Padisak, 2007) and morphologically based functional groupings (MBFGS, Kruk et al., 2010), according to which similar species are classified

together and they are expected to have, to a greater or lesser extent, common morphological or functional characteristics.

The study of phytoplankton in Bulgarian reservoirs was based mainly on the traditional approach related to the study of taxonomic structure and its seasonal succession. Research in this direction is significant in number, but results based on the modern environmental, functional and morphological concepts mentioned above are very rare. The relevance of the outlined issues and the lack of such studies have determined the purpose of the presented study, which has focused mainly on the implementation of these environmental approaches to better clarify the variations in the development of phytoplankton communities in large reservoirs that have long been used for aquaculture in the country.

## Materials and Methods

### Study area

Four of the surveyed reservoirs are located in southern Bulgaria (Dospat IBW 3155, Kardzhali IBW 1668, Zhreb-

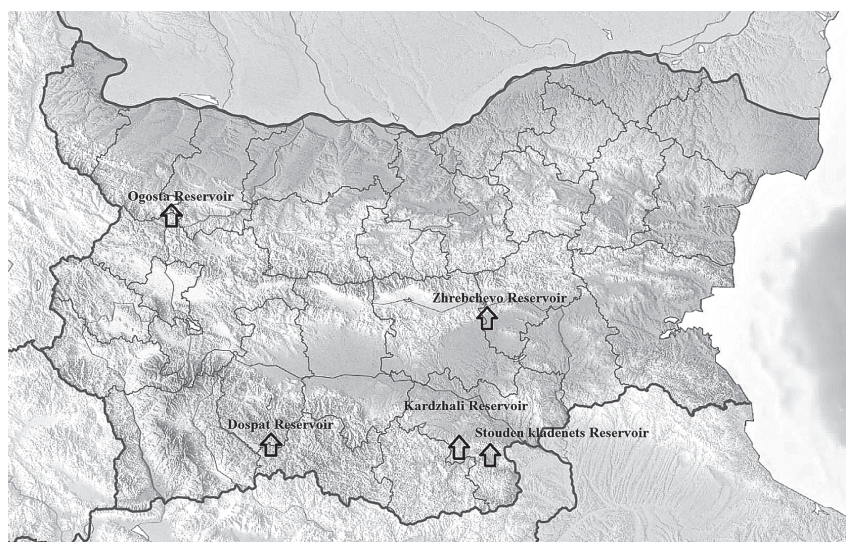


Fig. 1. Location of the studied reservoirs in Bulgaria

Table 1. Geographic locations and main morphometric features of the studied reservoirs

Reservoirs	DR	KR	ZR	SKR	OR
Altitude/m.a.s.l. (m)	1200	280	269	225	186
Length (km)	19	20	16	29	6.26
Width (km)	3	1.3	2.8	1.5	2.6
Max depth (m)	50	85	52	60	56
Water volume (m <sup>3</sup> )	497 x 10 <sup>6</sup>	449 x 10 <sup>6</sup>	400 x 10 <sup>6</sup>	387 x 10 <sup>6</sup>	506 x 10 <sup>6</sup>
Main source/river	Dospat	Arda	Tundzha	Arda	Ogosta
Built/year	1969	1957-1963	1966	1954-1957	1958-1986
Geographical coordinates	41° 38' 36" N 24° 9' 32" E	41° 38' 24" N 25° 22' 15" E	42° 36' 56" N 25° 51' 33" E	41° 38' 28" N 25° 32' 48" E	43° 22' 31" N 23° 10' 56" E

chevo IBW 2545 and Stouden Kladenets IBW 1763) and one (Ogosta IBW 1137) in the northern part of the country (Michev and Stoyneva, 2007, Figure 1). There are caged fish farms in the five studied reservoirs. The cultured species in reservoirs Kardzhali (KR), Stouden Kladenets (SKR), Zhrebchevo (ZR) and Ogosta (OR) include sturgeons (Acipenseridae), common carp (*Cyprinus carpio*) and wels (*Silurus glanis*) while in Dospat reservoir (DR) only rainbow trout (*Oncorhynchus mykiss*) is reared. The main morphometric characteristics of the reservoirs are presented on Table 1.

### Sampling and analysis

During the study a total 25 phytoplankton samples were taken from 5 large reservoirs used for aquaculture: Dospat (7), Zhrebchevo (4), Stouden Kladenets (4), Ogosta (2) and Kardzhali (8) in the period 2016-2017 from sampling sites located near the cage farms in the reservoirs (Figure 1). The water samples for analysis of phytoplankton were collected and processed by standard methods of fixation with forma-

lin to final concentration 4% and further sedimentation (ISO 5667-1:2006/AC:2007; ISO 5667-3:2003/AC:2007).

The analysis of phytoplankton was done on Bürker blood-counting chamber (Laugaste, 1974). The species composition was determined by light microscope (Carl Zeiss, Axioscope 2 plus) with magnification 400x using standard taxonomic literature with critical use of AlgaeBase (Guiry and Guiry, 2018). Diatoms were identified according to Cox (1996). The biomass was estimated by the method of stereometric approximations (Rott, 1981; Deisinger, 1984). The total biomass of each sample was assessed, defined as the amount of biomass of all species, summarized in separate taxonomic groups. Phytoplankton taxa were classified into phytoplankton functional classifications: functional groups (FGs, Reynolds et al., 2002; Padisak et al., 2009; Borics et al., 2016); morpho-functional groups (MFGs, Salmaso and Padisak, 2007); and morphologically based functional groups (MBFGs, Kruk et al., 2010). Functional classifications were defined for species that contributed at least 5%

**Table 2. List of identified phytoplankton species in five Bulgarian large reservoirs: Kardzhali (KR), Dospat (DR), Stouden Kladenets (SKR), Zhrebchevo (ZR) and Ogosta (OR)**

RESERVOIRS	KR	DR	ZR	SKR	OR
1	2	3	4	5	6
<b>Cyanoprokaryota</b>					
<i>Anabaena planctonica</i> Brunthaler	**				
<i>Anabaena</i> sp.	*	*	*		
<i>Anabaena sphaerica</i> Bornet & Flahault		**	*		
<i>Anabaenopsis arnoldii</i> Aptekar	*				
<i>Anathece clathrata</i> (W.West & G.S.West) Komárek, Kastovsky & Jezberová	**	**	*	*	
<i>Aphanizomenon flos-aquae</i> Ralfs ex Bornet & Flahault	**	*	**		*
<i>Aphanizomenon gracile</i> Lemmermann	**		*		
<i>Aphanocapsa delicatissima</i> West & G.S.West		*			*
<i>Aphanocapsa</i> sp.	*	*	*		
<i>Chroococcus minutus</i> (Kützing) Nägeli		*			
<i>Chroococcus turgidus</i> (Kützing) Nägeli	**		*	*	
<i>Dolichospermum spiroides</i> (Klebahn) Wacklin, L.Hoffmann & Komárek	**	*			
<i>Dolichospermum viguieri</i> (Denis & Frémy) Wacklin, L.Hoffmann & Komárek		**			
<i>Gloeotrichia echinulata</i> P.G.Richter		**		*	
<i>Limnococcus limneticus</i> (Lemmermann) Komárková, Jezberová, O.Komárek & Zapomelová	**	**	*	*	
<i>Merismopedia punctata</i> Meyen				*	
<i>Microcystis aeruginosa</i> (Kützing) Kützing	**			*	
<i>Microcystis</i> sp.	*				*
<i>Microcystis wesenbergii</i> (Komárek) Komárek ex Komárek	**				
<i>Oscillatoria</i> sp.			*		
<i>Planktolingbya limnetica</i> (Lemmermann) Komárková-Legnerová & Cronberg			**		
<i>Pseudanabaena mucicola</i> (Naumann & Huber-Pestalozzi) Schwabe	**				
<i>Snowella lacustris</i> (Chodat) Komárek & Hindák	*		*		*
<i>Woronichinia naegeliana</i> (Unger) Elenkin	**				
<b>Chlorophyta</b>					

Table 2. Continued

	1	2	3	4	5	6
<i>Ankyra judayi</i> (G.M.Smith) Fott		**			**	*
<i>Characium angustum</i> A.Braun			*		*	
<i>Chlorella vulgaris</i> Beyerinck [Beijerinck]						*
<i>Coelastrum astroideum</i> De Notaris					**	
<i>Coelastrum microporum</i> Nägeli		**	*	*		
<i>Coelastrum</i> sp.				*		**
<i>Coenochloris fottii</i> (Hindák) Tsarenko				*		
<i>Crucigenia quadrata</i> Morren						*
<i>Crucigenia</i> sp.				*	*	*
<i>Crucigeniella irregularis</i> (Wille) P.M.Tsarenko & D.M.John					*	*
<i>Crucigeniella pulchra</i> (West & G.S.West) Komárek				*		
<i>Desmodesmus bicaudatus</i> (Dedusenko) P.M.Tsarenko		*				
<i>Desmodesmus communis</i> (E.Hegewald) E.Hegewald		*		*		
<i>Dictyosphaerium simplex</i> Korshikov						*
<i>Eudorina elegans</i> Ehrenberg					*	
<i>Hariotina polychorda</i> (Korshikov) E.Hegewald				**	**	**
<i>Hariotina reticulata</i> P.A.Dangeard					*	*
<i>Lemmermannia triangularis</i> (Chodat) C.Bock & Krienitz						*
<i>Mucidosphaerium pulchellum</i> (H.C.Wood) C.Bock, Proschold & Krienitz		*	*	*		*
<i>Nephrocystium agardhianum</i> Nägeli		*	*			
<i>Oocystis lacustris</i> Chodat		**	*	**	**	*
<i>Pandorina morum</i> (O.F.Müller) Bory		*	**	**	**	**
<i>Pediastrum duplex</i> Meyen		*		*	*	*
<i>Pediastrum simplex</i> Meyen		*		*		*
<i>Planktosphaeria gelatinosa</i> G.M.Smith			*	*		*
<i>Radiococcus polycoecus</i> (Korshikov) I.Kostikov, T.Darienko, A.Lukesová & L.Hoffmann		*				
<i>Scenedesmus arcuatus</i> (Lemmermann) Lemmermann						*
<i>Scenedesmus bicaudatus</i> Dedusenko						*
<i>Scenedesmus</i> sp.				*		
<i>Schroederia spiralis</i> (Printz) Korshikov						*
<i>Sphaerocystis planctonica</i> (Korshikov) Bourrelly		*	*			
<i>Tetradesmus lagerheimii</i> M.J.Wynne & Guiry		*				
<i>Tetradesmus obliquus</i> (Turpin) M.J.Wynne		*			*	**
<i>Tetraedriella</i> sp.			*			
<i>Tetraëdron minimum</i> (A.Braun) Hansgirg		*		*	*	*
<i>Tetrastrum</i> sp.				*		
<b>Streptophyta</b>						
<i>Closterium aciculare</i> T.West		**		**	**	*
<i>Closterium acutum</i> Brébisson				**		*
<i>Cosmarium baileyi</i> Wolle			*			
<i>Cosmarium margaritifera</i> Meneghini ex Ralfs		**			*	**
<i>Cosmarium</i> sp.		**	*	*		*
<i>Elakatothrix gelatinosa</i> Wille		**		**	**	*
<i>Elakatothrix</i> sp.					*	
<i>Elakatothrix spirochroma</i> (Reverdin) Hindák				*		
<i>Staurastrum chaetoceras</i> (Schröder) G.M.Smith				*		
<i>Staurastrum gracile</i> Ralfs ex Ralfs				*		**
<i>Staurastrum pingue</i> var. <i>planctonicum</i> (Teiling) Coesel & Meersters		**	*	**	*	
<i>Staurastrum</i> sp.				*	*	*

Table 2. Continued

1	2	3	4	5	6
<i>Staurodesmus dejectus</i> (Brébisson) Teiling		**			
<i>Teilingia granulata</i> (J.Roy & Bisset) Bourrelly		*			
<b>Euglenophyta</b>					
<i>Euglena</i> sp.		*		*	
<i>Euglena viridis</i> (O.F.Müller) Ehrenberg			*		
<i>Phacus longicauda</i> (Ehrenberg) Dujardin			*		
<i>Phacus</i> sp.		*			
<i>Trachelomonas hispida</i> (Perty) F.Stein					*
<i>Trachelomonas nigra</i> Svirenko		*			
<i>Trachelomonas planctonica</i> Svirenko		*	*		
<i>Trachelomonas</i> sp.			*	*	
<i>Trachelomonas volvocina</i> (Ehrenberg) Ehrenberg			*	*	
<b>Pyrrhophyta</b>					
<i>Ceratium hirundinella</i> (O.F.Müller) Dujardin	*	*	**	**	
<b>Cryptophyta</b>					
<i>Cryptomonas</i> sp.	*				
<i>Rhodomonas lacustris</i> Pascher & Ruttner				*	
<i>Rhodomonas</i> sp.				*	
<b>Ochrophyta</b>					
<b>Synurophyceae</b>					
<i>Mallomonas</i> sp.	*	**	*	*	*
<i>Mallomonas acaroides</i> Zacharias				*	
<i>Mallomonas tonsurata</i> Teiling		**			
<b>Bacillariophyceae</b>					
<i>Achnanthes</i> sp.	*				
<i>Asterionella formosa</i> Hassall	**	*	*	*	*
<i>Aulacoseira granulata</i> (Ehrenberg) Simonsen	**	*	*		*
<i>Cocconeis pediculus</i> Ehrenberg	*			*	
<i>Cocconeis placentula</i> Ehrenberg	**	*	**	**	*
<i>Cocconeis</i> sp.		*			
<i>Cyclotella</i> sp.		*	*	*	*
<i>Cymatopleura solea</i> (Brébisson) W.Smith				*	
<i>Diatoma</i> sp.	*				
<i>Diatoma vulgare</i> Bory	*			*	
<i>Discostella stelligera</i> (Cleve & Grunow) Houk & Klee			**		
<i>Epithemia</i> sp.			*		
<i>Eunotia</i> sp.				*	
<i>Fragilaria acus</i> (Kützing) Lange-Bertalot			*	*	
<i>Fragilaria capucina</i> Desmazières			*		
<i>Fragilaria crotonensis</i> Kitton	**	**	**	**	
<i>Lindavia bodanica</i> (Eulenstein ex Grunow) T.Nakov, Guillory, Julius, Theriot & Alverson			*		
<i>Navicula</i> sp.		*	*	*	*
<i>Pantocsekiella comensis</i> (Grunow) K.T.Kiss & E.Ács					**
<i>Stephanodiscus astraea</i> (Kützing) Grunow			*		
<i>Stephanodiscus hantzschii</i> Grunow	*	*	**	**	**
<i>Tabellaria fenestrata</i> (Lyngbye) Kützing		*			
<i>Ulnaria ulna</i> (Nitzsch) Compère	*	*			**

\* presence, \*\* dominant species

of the mean phytoplankton biomass (Reynolds et al., 2002, Reynolds, 2006).

## Results

A total of 113 taxa of planktonic algae were found during the study of the five reservoirs: Cyanoprokaryota (24, 21.2%); Chlorophyta (36, 31.9%); Streptophyta (14, 12.4%); Pyrrophyta (1, 0.9%); Cryptophyta (3, 2.7%) and Ochrophyta (26, 23% Synurophyceae 3, 2.7%; Bacillariophyceae 23, 20.4%), forty-two of them were identified as dominant species (Table 2).

The most abundant phytoplankton species in the studied reservoirs were classified in eighteen functional groups (FGs): **A, B, C, D, E, F, G, H1, H2, J, K, Lo, M, MP, N, P, S1, X1**, given in Table 3 and Table 4. In all the studied reservoirs, the following functional groups contained the greatest variety of species: **H1, J, N, Lo, P** and **M**. Some of the dominant cyanoprokaryotes of codon **H1** in KR reservoir were: *Aphanizomenon flos-aquae* Ralfs ex Bornet & Flahault, *Aphanizomenon gracile* Lemmermann, and *Dolichospermum spiroides* (Klebban) Wacklin, L. Hoffmann & Komárek, and of codon **M**: *Microcystis aeruginosa* (Kützing) Kützing, *Microcystis wesenbergii* (Komárek) Komárek ex Komárek, known as potential producers of toxins. In the same reservoir there was also a great abundance of *Pseudanabaena mucicola* (Naumann & Huber-Pestalozzi) Schwabe and *Woronichinia naegeliana* (Unger) Elenkin (Table 3).

In high-altitude DR reservoir the blue-green algae were among the most abundant species: *Dolichospermum viguieri* (Denis & Frémy) Wacklin, L.Hoffmann & Komárek (**H1**) and *Gloeotrichia echinulata* P. G. Richter (**H2**), and in lowland ZR *Aphanizomenon flos-aquae* (**H1**) and *Planktolyngbya limnetica* (Lemmermann) Komárková-Legnerová & Cronberg (**S1**). From the green algae, *Oocystis lacustris* Chodat (**F**) was among the most abundant species in KR, ZR, and SKR, while *Pandorina morum* (O.F.Müller) Bory (**G**) was among the dominant species in DR, ZR, SKR and OR. *Ankyra judayi* (G.M.Smith) Fott of codon **X1** was among the most abundant in KR, SKR, and *Hariotina polychorda*

(Korshikov) E.Hegewald, of functional group **J**, in ZR, SKR and OR. Streptophytes *Closterium aciculare* T.West (**P**) and *Elakatothrix gelatinosa* Wille (**F**) dominated KR, ZR and SKR reservoirs. *Ceratium hirundinella* (O.F.Müller) Dujardin (**L<sub>0</sub>**) was among the most common species in ZR and SKR, and members of codon **E** *Mallomonas tonsurata* Teiling and *Mallomonas* sp. from Synurophyceae were part of the dominant complexes in DR.

Diatoms *Cocconeis placentula* Ehrenberg (**MP**) were among the most abundant species in KR, ZR and SKR reservoirs, *Fragilaria crotonensis* Kitton (**P**) dominated KR, DR, ZR and SKR, and the representative of the functional codon **D**, *Stephanodiscus hantzschii* Grunow, dominated ZR, SKR and OR reservoirs. Members of codons **P**: *Staurastrum pingue* var. *planctonicum* (Teiling) Coesel & Meersters and **N**: *Cosmarium margaritifera* Meneghini ex Ralfs, *Cosmarium* sp. and *Staurodesmus dejectus* (Brébisson) Teiling were among the most abundant species in all the studied reservoirs, except for SKR reservoir, while *Pandorina morum* (O.F.Müller) Bory of codon **G** was among the dominant species in all the reservoirs, except for KR reservoir. The member of functional group **K** *Anathece clathrata* (W.West & G.S.West) Komárek, Kastovsky & Jezberová was identified as one of the dominant species in KR and DR, and those of codons **A** (*Pantocsekiella comensis* (Grunow) K.T.Kiss & E.Ács) and **C** (*Asterionella formosa* Hassall) dominated OR and KR reservoirs.

According to the classification of Salmaso and Padišák (2007), there are fifteen identified morpho-functional groups (MBFs): **1b, 2a, 3b, 5a, 5b, 5c, 5e, 5d, 6a, 6b, 7a, 8a, 9b, 11a** and **11c** (Table 3, 4). The most common are: **5a, 5b, 5e, 6b, 8a** and **9b**. Large unicellular organisms of Conjugatophytes/chlorophytes (**8a**) were among the dominant species in most of the lowland reservoirs, KR, ZR, SKR and OR. The representatives of **9b** were also among the most abundant species in KR, ZR, SKR and OR reservoirs.

On the other hand, the members (Nostocales) of morpho-functional group **5e** were among the most common in KR and DR reservoirs. Large pennate diatoms (**6b**) were among the dominant species in KR, ZR and SKR reser-

**Table 3. List of identified groups based on different ecological approaches: FGs (Reynolds et al., 2002; Padišák et al., 2009; Borics et al., 2016), MFGs (Salmaso and Padišák, 2007) and MBFGs (Kruk et al., 2010)**

Reservoir	Ecological Groups		
	FGs	MFGs	MBFGs
KR	C, F, H1, J, K, Lo, M, MP, N, P, S1, X1	5a, 5b, 5c, 5e, 5d, 6a, 6b, 8a, 9b, 11c	I, III, IV, V, VI, VII
DR	E, G, H1, H2, K, Lo, P, N	2a, 3b, 5c, 5e, 6b, 8a	II, III, IV, V, VI, VII
ZR	B, D, F, G, H1, J, L <sub>0</sub> , MP, P, S1	1b, 3b, 5a, 6b, 7a, 8a, 9b, 11c	I, III, IV, V, VI, VII
SKR	D, F, G, J, L <sub>0</sub> , MP, P, X1	1b, 3b, 6b, 7a, 8a, 9b, 11c	I, IV, V, VI, VII
OR	A, D, G, J, N, P	3b, 6b, 7a, 8a, 9b, 11a	IV, V, VI

**Table 4. Morpho-functional classifications of phytoplankton in the investigated reservoirs according to: FGs (Reynolds et al., 2002; Padisák et al., 2009; Borics et al., 2016), MFGs (Salmaso and Padisák, 2007) and MBFGs (Kruk et al., 2010)**

FGs	MFGs	MFGs / morphology	MBFGs	MBFGs / morphology
		Colonies Cyanobacteria	<b>I</b>	Small organisms with high S/V
<b>Lo</b>	<b>5d</b>	Small Chroococcales colonies		
<b>X1, J, F</b>	<b>9b</b>	Small unicells (Chlorococcales)		
<b>E</b>	<b>2a</b>	Small Chrysophytes/Haptophytes	<b>II</b>	Small flagellated organisms with siliceous exoskeletal structures
<b>G</b>	<b>3b</b>	Colonial Phytomonadina		Large filaments with aerotopes
<b>H1, S1</b>	<b>5a</b>	Thin filaments (Oscillatoriales)	<b>III</b>	
<b>H1, H2, K</b>	<b>5e</b>	Nostocales		
<b>Lo</b>	<b>5c</b>	Large colonies, mostly non-vacuolated	<b>IV</b>	Organisms of medium size lacking specialized traits
		Chroococcales		
<b>P, N</b>	<b>8a</b>	Large unicells		
		(Conjugatophytes/Chlorophytes)		
<b>J</b>	<b>11a</b>	Chlorococcales (Naked colonies)		
<b>F</b>	<b>11c</b>	Other colonies		
<b>Lo</b>	<b>1b</b>	Large Dinophytes	<b>V</b>	Unicellular flagellates of medium to large size
<b>P</b>	<b>6a</b>	Large Centrics	<b>VI</b>	Non-flagellated organisms with siliceous exoskeletons
<b>C, D, MP, P</b>	<b>6b</b>	Large Pennates		
<b>A, D</b>	<b>7a</b>	Small Centrics		
	<b>5b</b>	Large vacuolated Chroococcales	<b>VII</b>	Large mucilaginous colonies
<b>M</b>				

voirs, while small centric diatoms (**7a**) were found in ZR, SKR and OR. The thin filamentous (Oscillatoriales) algae of morpho-functional group **5a** as well as the large vacuolated (Chroococcales) of group **5b** were among the representatives with the greatest variety of species in KR and ZR reservoirs. The members of MFGS **2a** and **1b**, small chrysophytes and large pyrrhophytes, were among the dominant species in DR, ZR and SKR reservoirs. Colonial Phytomonadina **3b** were found in all reservoirs except for KR. The large and small colonial species (Chroococcales) of **5c** and **5d** groups were dominant only in KR and DR reservoirs, while the large-centric diatoms of group **6a** were dominant only in KR reservoir. The members of group **11c** colonials dominated KR, ZR and SKR, while group **11a** (naked colonies Chlorococcales) were found only in OR (Table 3, 4).

By using the concept of Kruk et al. (2010), seven morphologically based functional groups (**MBFGs**) were estab-

lished: **I, II, III, IV, V, VI** and **VII** (Table 3, 4). Groups **III, IV, VI** and **VII** were presented with most species. The representatives of group **IV**, organisms of an average size, in which no specialized characteristics are available, were identified in KR, DR, ZR, SKR and OR. Non-flagellate organisms with silica exoskeleton of group **VI** were among the most common species in all the studied reservoirs with the exception of OR reservoir. The large gelatinous colonial organisms classified in group **VII** dominated KR, ZR and SKR reservoirs. The large filamentous (Nostocales/Oscillatoriales) of group **III** dominated KR, ZR and DR. The representatives of group **I**, small organisms (with high ratio of S/V) were among the dominant species in KR, ZR and SKR reservoirs, and small flagellates with silica exoskeletal structures of group **II** were among the dominants only in DR. Unicellular flagellates of medium to large size of group **V** were among the most abundant species in all the reservoirs studied (Table 3, 4).

## Discussion

In the study of phytoplankton, the application and comparison of various environmental classification systems, as well as the categorization of species in environmental groups have been widely used in recent years for monitoring and evaluation of the trophic state of different types of water basins (Salmaso et al., 2012; Izaguirre et al., 2012; Hu et al., 2013; Abonyi et al., 2014; Mihaljević et al., 2014; Zutinic et al., 2014; Salmaso et al., 2015; Török et al., 2016). This approach allows reduction of the number of variables in examining the ecological and trophic state of water basins (Hu et al., 2013). Phytoplankton is a huge and heterogeneous group of organisms, and this fact often hinders the development and interpretation of various environmental models. The classification of phytoplankton species with similar environmental characteristics facilitates the interpretation of data obtained from the monitoring of various freshwater ecosystems. Presently, there are three main environmental concepts in the study of phytoplankton communities: functional groups, morpho-functional groups and morphologically based functional groups (Hu et al., 2013).

According to the results of this phytoplankton study, different species of Cyanoprokaryota and Chlorophyta, Streptophyta and class Bacillariophyceae were dominant. According to the three morpho-functional concepts, the dominant groups of phytoplankton in the studied reservoirs were classified as follows: cyanoprokaryotes in six FGs (**H1**, **K**, **Lo**, **H2**, **M**, **S1**), five MFGs (**5a**, **5b**, **5c**, **5e**, **5d**) and three MBFGs (**III**, **IV**, **VII**), green algae in four FGs (**J**, **F**, **G**, **X1**), three MFGs (**9b**, **3b**, **11a**) and five MBFGs (**I**, **IV**, **V**, **VII**), streptophytes in three FGs (**P**, **N**, **F**), two MFGs (**8a**, **11c**) and two MBFGs (**IV**, **I**) and respectively diatoms in five FGs (**C**, **MP**, **D**, **P**, **A**), two MFGs (**6b**, **7a**) and one MBFGs (**VI**). Cyanoprokaryotes were among the dominants in three of the studied reservoirs (KR, DR and ZR), while some potentially toxic species were identified among them. The largest and longest functioning cage fish farms in Bulgaria are located in KR and DR reservoirs. The data of this study once again confirm the presence of species indicative of eutrophication, as well as of species known as potential producers of toxins in some of the largest reservoirs, such as KR and DR reservoirs, used for aquaculture production in the country.

The blue-green algae *Aphanizomenon flos-aquae* was reported among the dominants in Vacha reservoir (Belkinova et al., 2014), Kardzhali (Dochin and Iliev, in press), Dospat (Dochin and Stoyneva, 2014, 2016), Tsankov Kamak (Dochin and Ivanova, 2017) and Batak (Dochin et al., 2018). *Dolichospermum spiroides* was found in Kardzhali and Dospat reservoirs (Dochin and Stoyneva, 2014). According to

Reynolds et al. (2002) and Padisak et al. (2009), the members of codon **H1** *Aphanizomenon flos-aquae*, *Dolichospermum viguieri* and *Dolichospermum spiroides* are found in eutrophic, stratified and poor nutrient lakes, and according to MFGs and MBFGs concepts, they belong to the group of large filamentous species (**5a**; **5e**; **III**) Nostocales and Oscillatoriales (Salmaso and Padisak 2007; Kruk et al., 2010).

According to the concept of FGs, *Microcystis aeruginosa* (**M**) inhabits small to medium-sized eutrophic lakes, and according to MFGs and MBFGs, this same species belongs to the group of large (**5b**; **VII**) colonial Chroococcales (Reynolds et al., 2002; Padisak et al., 2009; Salmaso and Padisak, 2007; Kruk et al., 2010). *Gloeotrichia echinulata* (**H2**) is characteristic of mesotrophic, deep stratified lakes, with sufficient light availability, and according to the other two classifications, it belongs to the large colonial species (**5e**; **VII**) Chroococcales (Reynolds et al., 2002; Salmaso and Padisak, 2007; Padisak et al., 2009; Kruk et al., 2010). On the other hand, *Planktolyngbya limnetica* and *Pseudanabaena mucicola* (**S1**) are among cyanoprokaryotes that are well-adapted to low light environments, and according to the other two environmental classifications, they belong to the thin (**5a**; **IV**) filamentous algae (Reynolds et al., 2002; Salmaso and Padisak, 2007; Padisak et al., 2009; Kruk et al., 2010). The first species were abundant in 2015 in Kardzhali reservoir and the second ones were found in Ogosta and Koprinka reservoirs (Stoyanov et al., 2016; Dochin et al., 2017). *Woronichinia naegeliiana* (**Lo**; **5b**; **VII**), comparatively abundant in KR, was found to be among the dominant species in Batak reservoir (Dochin et al., 2018).

According to Padisak et al. (2009), the green *Oocystis lacustris* (**F**; **9b**; **VII**) found in KR, ZR and SKR reservoirs, inhabits deep mixed eutrophic lakes, and belongs to the large Chlorococcales (Salmaso and Padisak, 2007; Kruk et al., 2010). *Pandorina morum* (**G**; **3b**; **V**) found in abundance in DR, ZR, SKR and OR reservoirs, inhabits small, eutrophic and nutrient-rich lakes (Reynolds et al., 2002). According to Salmaso and Padisak (2007), the same species belong to the large unicellular organisms of Phytomonadina, and according to the theory of Kruk et al. (2010) – to the unicellular flagellates of medium to large size. The small-cell *Ankyra judayi* (**X1**; **9b**; **I**) dominant in KR, SKR reservoirs, is characteristic for shallow mixed waters (Reynolds et al., 2002; Salmaso and Padisak, 2007; Kruk et al., 2010). According to Belkinova et al. (2014) and Dochin and Iliev (in press), the last three species were among the dominant ones in Kardzhali reservoir. The green *Hariotina polychorda* (**J**; **9b**; **IV**), found in abundance in ZR, SKR and OR reservoirs, also inhabits shallow mixed waters (Salmaso and Padisak, 2007; Padisak et al., 2009; Kruk et al., 2010). The same species

were found in Zhrebchevo reservoir (Beshkova et al., 2014), while in 2015 their bloom was noted in Koprinka reservoir (reported by Dochin et al., 2017).

The large unicellular streptophytes *Closterium aciculare* (**P**; **8a**; **IV**) inhabits the epilimnion of stratified lakes, and *Elakatothrix gelatinosa* (**F**; **11c**; **I**) was dominant in KR, ZR, SKR reservoirs (Salmaso and Padisak, 2007; Padisak et al., 2009; Kruk et al., 2010). The last two species were among the dominant species in Kardzhali reservoir during the period 2015-2016 (Dochin and Iliev, in press). The large unicellular pyrrhophyte, *Ceratium hirundinella* (**L<sub>0</sub>**; **1b**; **V**), was among the most abundant species in ZR and SKR (Salmaso and Padisak, 2007; Kruk et al., 2010). These species inhabit deep and shallow, oligo- to eutrophic, medium and large lakes, according to Padisak et al. (2009), and was among the most abundant in 2015 in KR (Dochin and Iliev, in press).

The dominant species in DR, *Mallomonas tonsurata* and *Mallomonas* sp. (**E**; **2a**; **II**), belong to large flagellate from Synurophyceae, and according to the theory of the functional groups they usually inhabit small, shallow and poor trophic lakes (Reynolds et al., 2002; Salmaso and Padisak, 2007; Padisak et al., 2009; Kruk et al., 2010). According to Belkinova et al. (2014) *Mallomonas* sp. was in abundance in Toshkov Chark reservoir, and in Kardzhali reservoir according to Dochin and Iliev (in press). The diatom *Cocconeis placentula* (**MP**; **6b**; **VI**), found in KR, ZR and SKR reservoirs, which according to the concepts belongs to **MFGs** (large pennates) and **MBFGs** (non-flagellates with silica exoskeletons), often inhabit shallow and turbid lakes (Salmaso and Padisak, 2007; Padisak et al., 2009; Kruk et al., 2010). The above species were among the most abundant in 2015 in Kardzhali reservoir (Dochin and Iliev, in press).

The large pennate *Fragilaria crotonensis* (**P**; **6b**; **VI**), dominant in four of the five studied reservoirs (KR, DR, ZR and SKR), was also among the most abundant species in Kardzhali reservoir in 2015-2016, according to Dochin and Iliev (in press), and also in Krichim reservoir (Belkinova et al., 2014), Tsankov Kamak reservoir (Dochin and Ivanova, 2017) and Batak reservoir (Dochin et al., 2018). It is frequently found in the epilimnion of eutrophic, stratified lakes (Reynolds et al., 2002). According to this study, the small centric *Stephanodiscus hantzschii* (**D**; **7a**; **VI**), abundant in ZR, SKR and OR reservoirs, is typical for shallow and turbid waters (Reynolds et al., 2002; Salmaso and Padisak, 2007; Padisak et al., 2009; Kruk et al., 2010). It was among the most widespread species in 2015 in Kardzhali reservoir (Dochin and Iliev, in press).

According to Reynolds et al. (2002) streptophytes *Staurastrum pingue* var. *planctonicum* (**P**; **8a**; **IV**) inhabits poorly lit, stratified lakes with depletion of Si, while N members

*Cosmarium margaritiferrum*, *Cosmarium* sp. and *Staurodesmus dejectus* (**8a**; **IV**) are characteristic of stratified lakes with increased pH and deficiency of nutrients. The member of codon **A** *Pantocsekiella comensis* (**7a**; **VI**) inhabits lakes with increased pH and deficiency of nutrients, while *Asterionella formosa* (**C**; **6b**; **VI**) inhabit well-lit, stratified, low-carbon and silicon-depleted lakes (Reynolds et al., 2002). The representatives of codon **K** *Anathece clathrata* and codon **H1** *Anabaena planctonica* Brunnthaler (**5e**; **VII**) are found in shallow, nutrient-rich waters (Reynolds et al., 2002).

## Conclusions

In conclusion, according to the data obtained from the study, the representatives of Cyanoprokaryota, Chlorophyta, Streptophyta and Bacillariophyceae had the greatest contribution to the algal abundance in the five reservoirs included in the study, with a total of 113 taxa identified. According to the three studied morpho-functional approaches, the forty-two dominant species found were classified in 18 **FGs**, 15 **MFGs** and 7 **MBFGs**. In KR and DR reservoirs where the oldest and largest cage fish farms in the country are located, as well as in ZR reservoir, a large part of the dominant species were gelatinous colonial (**M**; **5b**; **VII**) and filamentous cyanoprokaryotes (**H1**; **5a**, **5e**, **III**), among which there are potentially toxic species which are well-known indicators of eutrophication. The proposed environmental concepts of functional groups (**FGs**), morpho-functional groups (**MFGs**) and morphologically based functional groupings (**MBFGs**) are, in our opinion, an important tool for understanding the seasonal changes in phytoplankton communities as well as for assessing the environmental status of different types of freshwater reservoirs. This study has shown that these environmental models can be successfully implemented in monitoring programs both for the assessment of changes and for the provision of more comprehensive information on the adaptations of phytoplankton communities in artificially modified reservoirs.

## References

- Abonyi, A., Leitão, M., Stanković, I., Borics, G., Várbíró, G., & Padisák, J. (2014). A large river (River Loire, France) survey to compare phytoplankton functional approaches: Do they display river zones in similar ways?. *Ecological Indicators*, 46, 11-22.
- Becker, V., Caputo, L., Ordóñez, J., Marcé, R., Armengol, J., Crossetti, L. O., & Huszar, V. L. (2010). Driving factors of the phytoplankton functional groups in a deep Mediterranean reservoir. *Water Research*, 44(11), 3345-3354.
- Becker, V., Huszar, V. L. M., & Crossetti, L. O. (2009). Respons-

- es of phytoplankton functional groups to the mixing regime in a deep subtropical reservoir. *Hydrobiologia*, 628(1), 137-151.
- Belkinova, D., Padisák, J., Gecheva, G., & Cheshmedjiev, S.** (2014). Phytoplankton based assessment of ecological status of Bulgarian lakes and comparison of metrics within the water framework directive. *Ecology and Environmental Research*, 12(1), 83-103.
- Beshkova, M., Kalchev, R., & Kalcheva, H.** (2014). Phytoplankton in the Zhrebchevo Reservoir (Central Bulgaria) before and after invasion of *Dreissena polymorpha* (Mollusca: Bivalvia). *Acta Zool. Bulg.*, 66(3), 399-409.
- Borics, G., Tóthmérész, B., Várбірó, G., Grigorszky, I., Czébel, A., & Görgényi, J.** (2016). Functional phytoplankton distribution in hypertrophic systems across water body size. *Hydrobiologia*, 764(1), 81-90.
- Cox, E. J.** (1996). Identification of freshwater diatoms from live material. Chapman & Hall, London, UK.
- Deisinger, G.** (1984). Leitfaden zur Bestimmung der planktischen Algen der Kärntner Seen und ihrer Biomasse. Karntner Institut für Seenforschung, Klagenfurt, Austria.
- Dochin, K. & Iliev, I.** Functional classification of phytoplankton in Kardzhali reservoir (Southeast Bulgaria) (In press).
- Dochin, K., & Ivanova, A.** (2017). The phytoplankton in Tsankov Kamak reservoir. *Zhivotnov'dni Nauki/Bulgarian Journal of Animal Husbandry*, 54(1), 35-49.
- Dochin, K., Ivanova, A., & Iliev, I.** (2017). The phytoplankton of Koprinka reservoir (Central Bulgaria): species composition and dynamics. *Journal of BioScience and Biotechnology*, 6(1), 73-82.
- Dochin, K., Kuneva, V., Ivanova, A., & Iliev, I.** (2018). Current state of phytoplankton in Batak reservoir (Southwestern Bulgaria). *Bulgarian Journal of Agricultural Science*, 24(4), 686-697.
- Dochin, K. T., & Stoyneva, M. P.** (2014). Effect of long-term cage fishfarming on the phytoplankton biodiversity in two large Bulgarian reservoirs. *Ber. nat.-med. Verein Innsbruck* (Berichte des naturwissenschaftlich-medizinischen Vereins), 99, 49-96.
- Dochin, K. T., & Stoyneva, M. P.** (2016). Phytoplankton of the Dospat Reservoir (Rhodopi Mts, Bulgaria) – indicator of negative trend in reservoir development due to long-term cage fish farming. *Ann. Univ. Sof., Fac. Biol.*, Book 2-Botany, 99, 47-60.
- Guiry, M. D., & Guiry, G. M.** (2018). AlgaeBase. World-wide electronic publication, National University of Ireland, Galway. <http://www.algaebase.org>.
- Hu, R., Han, B., & Naselli-Flores, L.** (2013). Comparing biological classifications of freshwater phytoplankton: a case study from South China. *Hydrobiologia*, 701(1), 219-233.
- Izaguirre, I., Allende, L., Escaray, R., Bustingorry, J., Pérez, G., & Tell, G.** (2012). Comparison of morpho-functional phytoplankton classifications in human-impacted shallow lakes with different stable states. *Hydrobiologia*, 1(698), 203-216.
- Kruk, C., Huszar, V. L., Peeters, E. T., Bonilla, S., Costa, L., Lüring, M., Reynolds, C.S. & Scheffer, M.** (2010). A morphological classification capturing functional variation in phytoplankton. *Freshwater Biology*, 55(3), 614-627.
- Laugaste, R.** (1974). Size and weight of the most widely distributed algae in the Tchudsko-Pskovsk and Vyrtysyarv lakes. *Gidrobiologicheskie issledovaniya*, 4, 7-23 (Ru).
- Michev, T. M., & Stoyneva, M. P.** (Eds.) (2007). Inventory of Bulgarian Wetlands and their Biodiversity. Part 1: Non-Lotic Wetlands. *Publ. House Elsi-M*, Sofia, 364 pp. + CD supplement.
- Mihaljević, M., Stević, F., Špoljarić, D., & Pfeiffer, T. Ž.** (2014). Application of morpho-functional classifications in the evaluation of phytoplankton changes in the Danube River. *Acta Zoologica Bulgarica*, 66(Suppl. 7), 153-158.
- Padisák, J., Crossetti, L. O., & Naselli-Flores, L.** (2009). Use and misuse in the application of the phytoplankton functional classification: a critical review with updates. *Hydrobiologia*, 621(1), 1-19.
- Reynolds, C. S., Huszar, V., Kruk, C., Naselli-Flores, L., & Melo, S.** (2002). Towards a functional classification of the freshwater phytoplankton. *Journal of Plankton Research*, 24(5), 417-428.
- Reynolds, C. S.** (2006). *The ecology of phytoplankton*. Cambridge University Press.
- Reynolds, C. S.** (1980). Phytoplankton Assemblages and Their Periodicity in Stratifying Lake Systems. *Holarctic Ecology*, 3(3), 141-159.
- Rott, E.** (1981). Some results from phytoplankton counting inter-calibrations. *Schweizerische Zeitschrift für Hydrologie*, 43(1), 34-62.
- Salmaso, N., & Padisák, J.** (2007). Morpho-functional groups and phytoplankton development in two deep lakes (Lake Garda, Italy and Lake Stechlin, Germany). *Hydrobiologia*, 578(1), 97-112.
- Salmaso, N., Naselli-Flores, L., & Padisák, J.** (2012). Impairing the largest and most productive forest on our planet: how do human activities impact phytoplankton?. *Hydrobiologia*, 698(1), 375-384.
- Salmaso, N., Naselli-Flores, L., & Padisák, J.** (2015). Functional classifications and their application in phytoplankton ecology. *Freshwater Biology*, 60(4), 603-619.
- Stoyanov, P., Teneva, I., Mladenov, R., & Belkinova, D.** (2016). Filamentous cyanoprokaryotes (Cyanoprokaryota/Cyanobacteria) in standing waters of Bulgaria: diversity and ecology. *Journal of BioScience & Biotechnology*, 5(1), 19-28.
- Stoyneva, M. P.** (2014). Contribution to the study of the biodiversity of hydro- and aerobic prokaryotic and eukaryotic algae in Bulgaria. Thesis for acquiring scientific degree "Doctor of Science", *Sofia University*, 825 pp.
- Stoyneva, M. P., Traykov, I., Uzunov, B., Zidarova, R., & Tosheva, A.** (2013). Perform phytoplankton monitoring in lakes as part of the National Surface Water Monitoring Program 2011-2013. Final report under Contract № 2081/01.09. 2011 with Executive Agency for Environmental Protection.
- Török, P., T-Krasznai, E., B-Béres, V., Bácsi, I., Borics, G., & Tóthmérész, B.** (2016). Functional diversity supports the biomass-diversity humped-back relationship in phytoplankton assemblages. *Functional Ecology*, 30(9), 1593-1602.
- Žutinić, P., Udovič, M. G., Borojević, K. K., Plenković-Moraj, A., & Padisák, J.** (2015). Morpho-functional classifications of phytoplankton assemblages of two deep karstic lakes. *Hydrobiologia*, 744(1), 317-317.