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# Long-term changes in phytoplankton composition in a large mountain reservoir used for cage aquaculture. A case study with Dospat Reservoir – Bulgaria

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#### Abstract

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Current research investigates changes in phytoplankton species composition over a seven-year period (2016–2022) and compares them with previous studies. Our objective is to focus on a three-year period (2019–2022) during which rainbow trout production decreases significantly, resulting in significant changes in algae species abundance and composition. During the same period, the species diversity of phytoplankton decreased two and a half times, and its biomass reached several times low-er levels than when the rainbow trout farm operated at full capacity. The number of identified cyanoprokaryotes has decreased twice, is not abundant and is almost not found among the dominant species. The adverse facts clearly confirm the impact of cage aquaculture on the eutrophication of Dospat Reservoir. We can predict with confidence that, in the next few years, the resumption of the production process will lead to a deterioration of the ecological status of the Dospat Reservoir, that will negatively affect the composition and structure of phytoplankton.

Keywords: cage fish farming; negative impact; lack of production; phytoplankton diversity; change; eutrophication

#### Introduction

Cage fish farming is an intensive method of rearing high density fish in natural aquatic systems, such as ponds, lakes, reservoirs, etc. (Chen & Zheng, 2005). The cage aquaculture significantly increases the risk of eutrophication. However, rearing methods, farm size, nature and volume of waste, depth and volume of lakes are very important (Phillips et al., 1985). The impact of this cultivation method includes an increase in turbidity and organic matter in the sediments and a decrease in dissolved oxygen, transparency and pH (Beveridge, 1984; Phillips et al., 1985; Pitta et al., 1999; Demir et al., 2001). Aquaculture has a clear impact on water quality as measured by phytoplankton abundance, but depends on the duration of fish rearing and the conditions specific to each reservoir (Miranda et al., 2016). Phytoplankton blooms caused by eutrophication of reservoirs are always among the major problems of aquatic systems (Huang et al., 2020). In aquaculture production, phytoplankton is a key contributor to primary production as a foundation for sustaining fish stocks and improving water quality. The study of factors controlling the composition of phytoplankton species and patterns of species diversity is important research objectives in fish farming (Zhang et al., 2021). Some authors hypothesize that in water bodies subjected to anthropogenic pressure, as water temperature and nutrient inputs increases, algal biomass increases, but their species diversity decreases (Wilk-Wozniak et al., 2013).

Under eutrophic conditions, despite high levels of biomass and phytoplankton density, the species diversity is very low (Toporowska et al., 2010). These variations in species composition, structure and quantitative distribution of phytoplankton can be used to evaluate eutrophication of reservoirs (Liu et al., 2017). One of the most appropriate indicators of changes due to nutrient loading is phytoplankton, because they respond quickly to these changes (Namin et al., 2021). In recent years, a significant number of studies on phytoplankton in Bulgarian reservoirs have been published, but research on the impact of cage fish farming on this community is still scarce. Therefore, the objective of the present study is focused on a period of three years (2019-2022) when the production of rainbow trout in the Dospat Reservoir is almost completely stopped. This lack of production is clearly linked to changes in the composition of the species and above all to the quantitative development of phytoplankton. The changes, which will be described in detail in the text below, are clear evidence of the impact of cage aquaculture as a major factor in the eutrophication of this reservoir.

#### **Material and Methods**

#### Study area and sampling

The study is conducted in a large mountain (1200 m.a.s.l.) Dospat Reservoir IBW 3155 (Michev & Stoyneva, 2007) (Figure 1, Table 1). The waters of this water body are mainly used for power generation and aquaculture. In the Dospat Reservoir is cultivated rainbow trout (*Oncorhynchus mykiss* Walbaum). There is build the largest and oldest rainbow trout cage fish farm in Bulgaria, which began over forty years ago. The average annual production of rainbow trout is around 800 tonnes. Fifty-two water samples for phytoplankton analysis are collected by Niskin-Type water sampler 5L model (Hydro-Bios Apparatebau GmbH, Germany). The phytoplankton samples are collected and processed by standard methods of fixation with formalin to final concentration 4% and further sedimentation (ISO5667-1: 2006/ AC:2007; ISO5667-3: 2003/AC: 2007). Microscope work has been done on Bürker chamber. The species composition is determined by light microscope (Carl Zeiss, Axioscope 2 plus) with magnification 400x using standard taxonomic literature with critical use of AlgaeBase (Guiry & Guiry, 2023). Diatoms are identified according to Cox (1996). The main counting unit is the cell, and the biomass is estimated by the method of stereometrical approximations (Rott, 1981; Deisinger, 1984). Counting is carried out individually (cell, filament or colony). The total biomass of each sample is assessed and it is defined as the amount of biomass of all species summarized in separate taxonomic groups. The similarity of the taxonomic composition of phytoplankton in the Dospat Reservoir during various study periods is calculated by the standard Sörensens' Similarity Index (SSI), according to the formula:

$$SSI = (2c/a + b),$$

where: a – number of species in period a, b – number of species of period b, and c – number of common species for periods a and b. Phytoplankton composition results for the period of 2016 to 2022 are compared to available species composi-



Table 1. Morphometric characteristics of Dospat Reservoir

Geographic coordinates	41°41′54″24°05′10″
Average height (m. a.s.l.)	1 200
Volume (m <sup>3</sup> )	449 248 693
Aquatory (m <sup>2</sup> )	22 099 371
Maximum length (m)	16 700
Average width (m)	683
Average depth (m)	20
Maximum depth (m)	50
Watershed basin (km <sup>2</sup> )	432.30
Retention time (days)	180

tion data published on the Dospat Reservoir for previous periods: for the period 1972–1975 by Naidenov & Saiz (1977) for the period 2010–2012 by Dochin & Stoyneva (2014; 2016) and for the period 2016–2017 by Dochin (2019).

#### Results

Fifty-two phytoplankton samples are analyzed in Dospat Reservoir from 2016 to 2022. A single sampling in 2016 recorded 20 planktonic algal taxa from 5 divisions: Cyanoprokaryota (3); Chlorophyta (4); Streptohyta (4); Euglenophyta (1); and Ochrophyta (8). The most abundant in cages are Staurodesmus dejectus (Brébisson) Teiling (56.82%) and Stephanodiscus hantzschii Grunow (33.22%) from Ochrophyta. The estimated biomass is 1.144 mg.L-1. In 2017, in 6 samplings from two sites, 29 taxa from 6 groups are found: Cyanoprokaryota (10); Chlorophyta (5); Streptohyta (4); Euglenophyta (4); Pyrrhophyta (1) and Ochrophyta (5). In the cages 24 taxa from 6 divisions found: Cyanoprokaryota (7); Chlorophyta (4); Streptohyta (4); Euglenophyta (3); Pyrrhophyta (1) and Ochrophyta (5). Near the wall are identified 19 taxa from 6 phytoplankton groups: Cyanoprokaryota (7); Chlorophyta (4); Streptohyta (2); Euglenophyta (2); Pyrrhophyta (1) and Ochrophyta (3). In August, a bloom of the cyanoprokaryote Dolichospermum viguieri (Denis & Frémy) Wacklin, L.Hoffmann & Komárek(100%) is recorded in the cages, with its biomass increasing to 13.848 mg.L<sup>-1</sup>. In the same site in late August, the above species is again most abundant, but with five times lower (2.721 mg.L<sup>-1</sup>) biomass. A change in the dominant algal species observed in the cages in early September. The most abundant are blue-green Gloeotrichia echinulata P.G.Richter(41.6%) and the diatom Fragilaria crotonensis Kitton (26.62%). In the wall, dominant species are colonial members of the Chlorophyta Pandorina morum (O.F.Müller) Bory(47.92%) and Sphaerocystis planctonica (Korshikov) Bourrelly (32.47%). In the wall, biomass values reached 2.88 mg.L-1. The mean value of biomass obtained from four samples in August and September in the cages is 4.970 mg.L<sup>-1</sup>, while in two samples at the wall in September this value is 2.445 mg.L<sup>-1</sup>. In 2017, the average value recorded for biomass is 3.708 mg.L<sup>-1</sup>. The only sample collected in 2018 in the cages is identified 10 taxa from 5 divisions: Cyanoprokaryota (1); Chlorophyta (4); Streptohyta (1); Pyrrhophyta (1) and Ochrophyta (2). The most abundant are *F. crotonensis*(51.6%), *Cosmarium* sp. (35.2%) and *Limnococcus limneticus* (Lemmermann) Komárková, Jezberová, O.Komárek & Zapomelová (13.4%). The biomass during the period is 1.385 mg.L<sup>-1</sup>.

Eight phytoplankton samples from two sites are collected in 2019. In June-September 2019, 40 taxa from 6 divisions are identified: Cyanoprokaryota (5); Chlorophyta (12); Streptohyta (5); Euglenophyta (1); Pyrrhophyta (2) and Ochrophyta (15). On the site near to the wall are found 29 taxa from 5 groups: Cyanoprokaryota (2); Chlorophyta (5); Streptohyta (1) and Ochrophyta (4). While in the cages are identified 31 taxa from 6 divisions: Cyanoprokaryota (1); Chlorophyta (2); Streptohyta (2); Pyrrhophyta (1) and Ochrophyta (2). The colonial green alga Sphaerocystis schroteri Chodat (65.7-92.9%) dominates in June. The diatom F. crotonensis (77-82.3%) is the most abundant in July and August (46.2-89.5%). Without significant change in September 2019 again dominated F. crotonensis (51-60.3%). The biomass varies from 0.194 mg.L<sup>-1</sup> in September in the reservoir wall to 3.572 mg.L<sup>-1</sup> in August in the cages. In 2019, the mean phytoplankton biomass in the wall is 0.502 mg.L<sup>-1</sup>, while in the fish farm is 1.353 mg.L<sup>-1</sup>. The mean algal biomass during the year is 0.928mg.L<sup>-1</sup>.

Sixty phytoplankton samples are collected from four sites in 2020. A total of 24 taxa from 6 divisions are identified: Cyanoprokaryota (4); Chlorophyta (9); Streptohyta (1); Euglenophyta (1); Pyrrhophyta (2) and Ochrophyta (7). In the wall are identified 18 taxa from 5 divisions: Cyanoprokaryota (3); Chlorophyta (7); Streptohyta (1) and Ochrophyta (2). In the middle part are identified 18 taxa from 5 divisions: Cyanoprokaryota (1); Chlorophyta (6); Euglenophyta (1); Pyrrhophyta (2) and Ochrophyta (5). In the cages are found 14 taxa from 4 groups: Cyanoprokaryota (2); Chlorophyta (6); Pyrrhophyta (1) and Ochrophyta (5). On the tail, 12 taxa from 5 groups are recorded: Cyanoprokaryota (3); Chlorophyta (3); Streptophyta (2); Pyrrhophyta (1) and Ochrophyta (3). The diatoms F. crotonensis (33.5-52.1%) and Asterionella formosa Hassall (46-62.8%) dominate late May. Different dominant algal taxa were identified in all sites in July, 2020. In the wall, the dominant are Characium sp. (40%), Pseudospharocystis cf. lacustris (47.4%) in the middle part, in cages *Willea irregularis* (Wille) Schmidle (57.7%), while in the tail F. crotonensis (47.4%).

The diatoms *Stephanodiscus astraea* (Kützing) Grunow (42.3%) dominates the wall in August, and *F. crotonen*-

sis (47.4%) the mid-section. Eudorina elegans Ehrenberg (56.5%) is most abundant in the cages, and in the tail co-dominated *F. crotonensis* (36.2%) and *S.schroteri* (33%). In October 2020 in the wall, the most abundant is *Tabellaria fenestrata* (Lyngbye) Kützing (75.4%), and in the middle part *A. formosa* (70.8%). In the cages the last species (49%) co-dominates with *Gymnodinium uberrimum* (G.J.Allman) Kofoid & Swezy (32.4%), whereas in the tail the most massive is *A. formosa* (88.2%). The biomass in 2020 varies widely from 0.0238 mg.L<sup>-1</sup> in the tail in October to 2.099 mg.L<sup>-1</sup> in the cages in August. The mean value for the biomass in the wall is 0.402 mg.L<sup>-1</sup>, in the middle part – 0.707 mg.L<sup>-1</sup>, to the cages – 0.773 mg.L<sup>-1</sup> and to the tail – 0.709 mg.L<sup>-1</sup>. The estimated average phytoplankton biomass in 2020 is 0.647 mg.L<sup>-1</sup>.

In 2021, eight samples from two sites are analyzed in the reservoir. Only 16 taxa from 6 divisions are identified: Cyanoprokaryota (5); Chlorophyta (3); Streptohyta (1); Cryptophyta (1); Pyrrhophyta (1) and Ochrophyta (5). In the site near to the wall are identified 12 taxa from 5 groups: Cyanoprokaryota (2); Chlorophyta (3); Streptohyta (1); Cryptophyta (1) and Ochrophyta (5). In the cages found 10 taxa from 4 divisions: Cyanoprokaryota (4); Chlorophyta (3); Pyrrhophyta (1) and Ochrophyta (2). In June, in the wall, the most abundant are Cyclotella sp. (40.2%) and Cocconeis sp. (29.1%). Asterionella formosa (45.9%) and W. irregularis (26.7%) are common in the cages in June. On the wall in July dominated T. fenestrata (88.7%), in the fish farm the last (75.8%) codominated together with G.uberrimum (24.2%). In August, in the wall the most common are *Characium* sp. (27.9%), W. irregularis (24.4%) and unidentified Chlorococcales (27.4%). While in the cages, unidentified Chlorococcales (49%) and T. fenestrata (27.5%). Blue-green Dolichospermum planctonicum (Brunnthaler) Wacklin, L.Hoffmann & Komárek (39.8%) and diatom A.formosa (35.4%) are ubiquitous in the wall in September. The fish farm is dominated by D. planctonicum and G. uberrimum. The biomass values vary from 0.135 mg.L<sup>-1</sup> to 1.984 mg.L<sup>-1</sup> at the wall. In 2021 the estimated average value is 0.735 mg.L<sup>-1</sup>, as in the wall it is 0.824 mg.L<sup>-1</sup>, and for cages is 0.779 mg.L<sup>-1</sup>.

In 2022, 12 phytoplankton samples collected from 3 sites. A total 37 taxa from 7 divisions are identified: Cyanoprokaryota (3); Chlorophyta (12); Streptohyta (5); Euglenophyta (3); Pyrrhophyta (2); Cryptophyta (1) and Ochrophyta (9). In the wall 20 taxa from 6 groups found: Cyanoprokaryota (1); Chlorophyta (8); Streptohyta (3); Euglenophyta (1); Pyrrhophyta (1)and Ochrophyta (6). In the cages identified 23 taxa from 7 divisions: Cyanoprokaryota (1); Chlorophyta (8); Streptohyta (1); Euglenophyta (3); Pyrrhophyta (2); Cryptophyta (1) and Ochrophyta (7). Twenty-two taxa from 6 divisions found in the tail: Cyanoprokaryota (3); Chlorophyta (6); Streptohyta (3); Euglenophyta (1); Pyrrhophyta (2) and Ochrophyta (7). In June, in the wall dominates the green algae *Sphaerocystis* sp. (37.5%) and *Monoraphidium contortum* (Thuret) Komárková-Legnerová (31%). During the same time in the cages the most massive is *Trachelomonas planctonica* Svirenko (81.3%). *Sphaerocystis* sp. (49.2%) and *Elakatothrix lacustris* Korshikov (46%) are most abundant in the tail. In July in the wall, the most abundant is *Mallomonas acaroides* Zacharias (56.7%), in the cages the last species (25.8%) co-dominates together with a *T. fenestrata* (39.4%).

Whereas the tail part is dominated by Ceratium hirundinella (O.F.Müller) Dujardin (44.1%) and F. crotonensis (22.9%). In September in the wall, the most common are T. fenestrata (46.3%) and S. dejectus (12.2%), T. fenestrata (68.7%-75.3%) are widely dominant in the cages and the tail. In October in the wall, the most common are diatoms T. fenestrata (57.1%) and F. crotonensis (14.9%), whereas in cages T. fenestrata (54.8%) and C. hirundinella (24.8%). In the tail, the last two species again co-dominated with a percentage contribution to phytoplankton biomass of respectively (42.1%) and (41.9%). A bloom of the last two species is recorded at the same site in October, with total biomass reaching a value of 14.094 mg.L<sup>-1</sup>. In 2022, the mean reported biomass at the wall is 2.095 mg.L-1, and for the cages and tail, respectively, 2.823 mg.L<sup>-1</sup> and 4.726 mg.L<sup>-1</sup>. The mean calculated biomass in 2022 is 3.215 mg.L<sup>-1</sup>.

Fifty years ago, 45 algae taxa found in phytoplankton research and 131 were registered in the 2010–2012 period (Naidenov & Saiz, 1977; Dochin & Stoyneva, 2014; Dochin, 2015). In the present study, 95 phytoplankton taxa are identified in Dospat Reservoir (Table 2). When comparing the similarity of species between the periods 1972–1975 and 2016-2022, there are 20 common species. The greatest species similarity found (with only one species common to both periods) in the division Cryptophyta (1.0), classes Bacillariophyceae (0.47) and Synurophyceae (0.4), as well as in the divisions Pyrrhophyta (0.4) and Chlorophyta (0.39 Figure 2).

With the greatest number of common species for the two study periods 10 in number, are the members of the class Bacillariophyceae. Overall, for all groups, the Sørensen species similarity coefficient is 0.35. The same comparison between 2010–2012 and 2016–2022 showed 33 common taxa, but the value of the Sørensen's species similarity coefficient is only 0.19. The greatest species similarity found in groups Cyanoprokaryota (0.41), Bacillariophyceae (0.39) and Streptophyta (0.38). With the highest number of similar species (15), again diatoms (Figure 3). During the same period, no common species were observed in four groups of algae (Euglenophyta,



Fig. 2. Sörensens' Similarity Index (SSI) between different taxonomic groups of algae, calculated by comparing the periods 1972–1975 with 2016–2022





Cryptophyta, Synurophyceae and Chrysophyceae). According to Dochin & Stoyneva (2014), the common species when comparing 1972–1975 with 2010–2012 is 22 and the species similarity by Sørensen's coefficient is only 0.25.

#### Discussion

Over the last forty years, Dospat Reservoir has been used for rearing rainbow trout. In recent years, a three-year period is clearly evident, during which aquaculture production is reduced significantly or almost totally absent, and that is exactly the period from 2019–2022. Considering data from previous years published by Dochin & Stoyneva (2014; 2016) on the impact of cage fish farming on the species composition of phytoplankton, the above-mentioned period in which there is an almost complete absence of fish farming stands out as different in terms of phytoplankton development in the reservoir. In 2016 *S. hantzschii* together with *S. dejectus* are among the dominant species. The first is found in our previous studies together with *S. astraea* (Dochin & Stoyneva, 2014; 2016; Dochin, 2019). According to Borics et al. (2007), members of the genus *Stephanodiscus* are common in small to medium-sized mesotrophic lakes, while others report them in eutrophic small to medium-sized lakes (Salmaso, 2002). Reynolds et al. (2002), note that members of the genus *Staurodesmus* are common in nutrient-poor stratified lakes.

What we do know is that last year there was a major production of trout in the Dospat Reservoir in 2017. This year, the biomass of cyanobacteria has increased considerably. In August, a bloom of *D.viguieri* is recorded, whose absolute biomass reached 13.848 mg.L<sup>-1</sup>. In September the last species is replaced by G. echinulata (Dochin, 2019). These two blue-green algae have never been observed in the Dospat Reservoir. These species were not observed in subsequent years of this study. Dolichospermum viguieri belongs to the common species in stratified eutrophic lakes (Moustaka-Gouni et al., 2007), whereas G. echinulata is ubiquitous in mesotrophic deep stratified lakes (Reynolds et al., 2002). The intense blooms of cyanobacteria, indicators of eutrophication, recorded during the year are clear evidence of the negative impact of cage fish farming on phytoplankton. In the same year, 10 taxa (39%) of the identified species belonged to the Cyanoprokaryota division, and the mean phytoplankton biomass for the year is 3.708 mg.L<sup>-1</sup>. For comparison, the mean phytoplankton biomass values for the 1970s and 2010-2012 are 0.7 and 1.35 mg.L<sup>-1</sup>, respectively (Naidenov & Saiz, 1977; Dochin & Stoyneva, 2016). The nearly triple increase in phytoplankton biomass in 2017 confirms once again the negative impact of cage aquaculture that we found (Dochin & Stoyneva, 2014; 2016).

In the single sampling in 2018, with the exception of *L. limneticus*, cyanoprokaryotes were not observed in dominant species, and the biomass is 1.385 mg.L<sup>-1</sup>. The above are part of the dominant species in 2010–2012 (Dochin & Stoyneva, 2014). According to Moura et al. (2007) it is widely distributed in shallow eutrophic waters. Similar to previous studies, *F. crotonensis* and *Cosmarium* sp. are among the most common in 2018 (Naidenov & Saiz, 1977; Dochin & Stoyneva, 2016). *Cosmarium* sp. is recorded in species abundant in mixed shallow waters (Kruk et al., 2002). The last reported taxa are among the most abundant in the 1970s and 2018, whereas in

2010-2012 they are common (Naidenov & Saiz, 1977; Dochin & Stoyneva, 2014). In 2019, when fish production has almost completely stopped, 40 algae are identified, but, in contrast to the high species abundance, the mean biomass is 0.928 mg.L<sup>-</sup> <sup>1</sup>, whereas the same in cages (1.353 mg.L<sup>-1</sup>) is two and a half times higher (0.502 mg.L<sup>-1</sup>) than that of the wall. No bluegreen algae were found among the dominants this year. Twenty-nine taxa are identified on the wall and thirty-one in the cages. Among them, only 5 taxa (12.5%) are members of the Cyanoprokaryota. In comparison with a total of 45 taxa identified in the 1970s, no cyanobacteria found in the 1970s, and 16 are registered in 2010-2012 (Naidenov & Saiz, 1977; Dochin & Stoyneva, 2014). Colonial green S. schroteri dominating in June, first recorded in 2019. The species occurs in small, shallower, polymictic waters (Kruk et al., 2002). Compared to 2017, the species abundance of algae is higher, but the average biomass is almost four times lower, confirming the impact of trout production on phytoplankton.

In 2020, as in the previous year, the exploitation of the fish farm is almost completely stopped. During the period, the number of identified taxa is 24. Together with the usual *F. crotonensis* and *A. formosa*, among the dominant species found for the first time *Pseudosphaerocystis* cf. *lacustris*. According Naselli-Flores & Barone (2003) this algae occurs in meso- and eutrophic reservoirs. *Fragilaria crotonensis* and *A. formosa* are reported among the dominant species in large, deep sub-alpine lakes with long reneval time periods (Salmaso, 2002). In the this study, the first species is not among the most abundant only in 2021, and the second is only registered in 2020 and 2021. *Fragilaria crotonensis* and *A. formosa* are consistently among the dominant algae in all previous survey years (Naidenov & Saiz, 1977; Dochin & Stoyneva, 2014; 2016; Dochin, 2019).

Among the four identified cyanobacteria (16.7%), no members entered the dominant phytoplankton complexes. In October 2020 similar to the study by Dochin & Stoyneva (2014; 2016) among the dominant species is identified G. uberrimum and T. fenestrata. Medium sized lakes of various trophic levels are inhabited by large algae such as G. uberrimum (Niesel et al., 2007). In the epilimnion of stratified lakes, where layer mixing is a fact, species such as T. fenestrata are among the abundant species (Dokulil & Teubner, 2003). The dominant in September 2017 P. morum and in August 2020 E. elegans is species inhabiting nutrient-rich waters (Huszar et al., 2003; Anneville et al., 2005). The last species are identified in the early 1970s and are also among the dominants during the period 2010-2012 (Naidenov & Saiz, 1977; Dochin & Stoyneva, 2014). The current year recorded the lowest mean phytoplankton biomass (0.647 mg.L<sup>-1</sup>) during the study. In 2020, the differences in average biomass at the different sites are negligible. In the same year, the species abundance decreases by 40% and the average biomass values continued to decline.

Only sixteen algae from six groups found in 2021. Together with the summer-dominant A. formosa and T. fenestrata in September, the most abundant are D. planctonicum and G. uberrimum. The first is recorded for the first time and only in the year 2021 as a dominant species in the Dospat Reservoir. It is important to note that this species is only identified in mid-September, but its abundance and biomass are not high. Several authors indicate D. planctonicum among the common species in shallow eutrophic lakes (Kruk et al., 2002; Crossetti & Bicudo, 2008a; 2008b). According to some researchers, the most common in 2021 Cyclotella sp. inhabits clear, deep lakes with an increasing tendency of pH (Reynolds et al., 2002; Soares et al., 2007). Whereas another abundant alga of the year, Cocconeis sp., is reported in turbid and shallow waters (Moura et al., 2007). The last two are found in the early 1970s, as in our previous research, but are not among the dominant species (Naidenov & Saiz 1977; Dochin & Stoyneva 2014; Dochin, 2019). Mean algal biomass value (0.735 mg.L<sup>-1</sup>) during the year are again significantly lower than in the years when the fish farm operated at full capacity. During the study, the differences between the mean phytoplankton biomass of the two sites are insignificant. The decline in species abundance and biomass for the previous two years continues in 2021.

In 2022, the species richness of phytoplankton increases to 37 taxa. According to our information, production is gradually resuming in the current year, and breeding of rainbow trout spawning material begins in the cage farm. During the summer months in the wall and in the cages, the most abundant species include T. planctonica, M. acaroides, whereas in the tail are abundant C. hirundinella, F. crotonensis and E. gelatinosa. In September, T. fenestrata and S. dejectus predominate, while in October, intense blooms of C. hirundinella and T. fenestrata are recorded. The euglenic T. planctonica is common in shallow meso- or eutrophic ponds (Padisák et al., 2003a). Ceratium hirundinella inhabits oligo to eutrophic, both deep and shallow water bodies of various sizes (Padisák & Reynolds, 1998; Salmaso, 2002). This species is only identified in 2022, but has a huge impact on algal biomass. Contrary to that, it was identified in the early 1970s and in 2016-2017, but is not dominant (Naidenov & Saiz, 1977; Dochin, 2019).

*Tabellaria fenestrata* abundant in 2021 and especially in 2022 is registered during 2010–2012 (Dochin & Stoyneva, 2014; 2016). It is a common species in clean oligotrophic lakes (Huszar et al., 2003). Also abundant in 2022, *M. contortum*, *M. acaroides* and *Elakatothrix lacustris* occur in nutrient-poor waters (Mazzeo et al., 2003). *Monoraphidium contortum*, first

discovered in a reservoir in 2022. Antenucci et al. (2005) indicate *Elakatotrix* sp. as a common in reservoirs with large temporal variations in the hydrological regime, while others refer him to pure deep mixed mesotrophic or eutrophic ponds (Reynolds et al., 2002). Due to the blooms, the mean biomass estimated (4.726 mg.L<sup>-1</sup>) at the tail is twice higher compared to those at the cages (2.823 mg.L<sup>-1</sup>) and the wall (2.095 mg.L<sup>-1</sup>). The mean biomass value calculated in 2022 is very close to what it was in 2017. In 2022, species abundance increases in the direction from the wall to the tail. The same can be said about the average biomass of phytoplankton. In most cases it is the normal horizontal distribution of phytoplankton in reservoirs. This latest result contrasts with the reverse distribution we previously found (Dochin & Stoyneva, 2016).

#### Conclusions

Finally, the presented study describes the changes in phytoplankton species composition for a period between

2016–2022, and compares them to periods fifty years ago (1972-1975) and ten years ago (2010-2012) in Dospat Reservoir. The investigation focused on a three-year period (2019-2022) during which, to our knowledge, fish production decreased and nearly ceased. We found important changes in the abundance and composition of algal species. The species diversity of phytoplankton decreases two and a half times and mean biomass also reached levels several times lower compared to periods when the trout production is operating at full capacity. During the same period, the number of found cyanobacteria species decreased twice, with low biomass values and almost no species found among the dominants. All the data provided clearly confirm the impact of cage aquaculture on the eutrophication processes of Dospat Reservoir. Therefore it can be predicted that the initiation of intensive trout production in the coming years will lead to a deterioration of the ecological status of the Dospat Reservoir, especially as refering to phytoplankton.

Table 2. List of identified phytoplankton species in Dospat Reservoir during different periods (acc. To Naidenow & Saiz, 1975; acc. to Dochin & Stoyneva, 2014; 2016; acc. to Dochin, 2019; 2016–2022 – data from present study). \*\*dominant species: \*occurrence

Taxon	Periods		
	1972-1975	2010-2012	2016-2022
Cyanoprokaryota			
Anabaena sp.		*	*
Anabaena sphaerica Bornet & Flahault		*	
Anabaenopsis sp.		*	**
Anathece clathrata (West & G.S.West) Komárek, Kastovsky & Jezberová			*
Aphanizomenon flos-aquae Ralfs ex Bornet & Flahault		**	*
Aphanocapsa delicatissima West & G.S.West			*
Aphanocapsa spp.		*	*
<i>Aphanothece</i> sp.			**
Calothrix sp.		*	
Chroococcus minutus (Kützing) Nägeli			*
Chroococcus turgidus (Kützing) Nägeli		*	**
Coelosphaerium kuetzingianum Nägeli		*	
Dolichospermum flos-aquae (Bornet & Flahault) P.Wacklin, L.Hoffmann & Komárek		*	
Dolichospermum planctonicum (Brunnthaler) Wacklin, L.Hoffmann & Komárek			**
Dolichospermum spiroides (Klebhan) Wacklin, L.Hoffmann & Komárek		*	*
Dolichospermum viguieri (Denis & Frémy) Wacklin, L.Hoffmann & Komárek			**
Gloeotrichia echinulata P.G.Richter			**
Gloeotrichia sp.			**
Limnococcus limneticus (Lemmermann) Komárková, Jezberová, O.Komárek & Zapomelová		**	**
Microcystis aeruginosa (Kützing) Kützing		*	
Noctoc sp.		*	
Oscillatoria limosa C.Agardh ex Gomont		**	
Phormidium sp.		*	*
Planktolyngbya sp.			*

Taxon	Periods			
	1972-1975	2010-2012	2016-2022	
Planktothrix agardhii (Gomont) Anagnostidis & Komárek		*		
Planktothrix rubescens (De Candolle ex Gomont) Anagnostidis & Komárek		**		
Snowella sp.			*	
Spirulina major Kützing ex Gomont		*		
Chrlorophyta				
Actinastrum hantzschii Lagerheim		*		
Ankyra judayi (G.M.Smith) Fott			*	
Characium angustum A.Braun			*	
Characium sp.			**	
Chlamydomonas simplex Pascher			*	
Coelastrum microporum Nägeli			*	
Coelastrum sp.			*	
Coelastrum sphaericum Nägeli		*		
Coenochloris sp.			*	
Coenococcus planctonicus Korshikov			*	
Coenococcus sp.			*	
Comasiella arcuata var. platydisca (G.M.Smith) E.Hegewald & M.Wolf	*			
Crucigenia tetreapedia (Kirchner) Kuntze	*		*	
Crucigeniella irregularis (Wille) P.M.Tsarenko & D.M.John		*	**	
Desmodesmus communis (E.Hegewald) E.Hegewald	*	*	*	
Enallax acutiformis (Schröder) Hindák		**		
Eudorina elegans Ehrenberg	*	**	**	
Golenkinia radiata Chodat		*		
Koliella sp.		*		
Korshikoviella cf. limnetica			*	
Korshikoviella sp.			*	
Micractinium pusillum Fresenius		*		
Monactinus simplex (Meven) Corda		*		
Monoraphidium contortum (Thuret) Komárková-Legnerová			**	
Mucidosphaerium nulchellum (H.C.Wood) C.Bock, Proschold & Krienitz	*	*	*	
Nephrocytium agardhianum Nägeli			*	
Oedogonium sp.		*		
Oocvstidium ovale Korshikov		**		
Oocystis borgei J.W.Snow		*	*	
Oocystis lacustris Chodat			*	
Oocystis sp.	*			
Pandorina morum (O.F.Müller) Bory		**	**	
Pediastrum duplex Meyen		*		
Planktosphaeria gelatinosa G.M.Smith			**	
Planktosphaeria sp.			**	
Pseudosphaerocystis cf lacustris			**	
Scenedesmus acuminatus var elongatus G M Smith		*		
Sphaerocystis schroeteri Chodat			**	
Sphaerocystis planctonica (Korshikov) Bourrelly			**	
Sphaerocystis sp			**	
Stichococcus sp			**	
Site in the set of the	1	1	1	

Taxon	Periods			
	1972-1975	2010-2012	2016-2022	
Tetradesmus lagerheimii var. biseriatus (Reinhard) Taskin & Alp	*	*		
Tetradesmus obliquus (Turpin) M.J.Wynne		*		
Tetraedriella sp.			*	
Tetrastrum glabrum (Y.V.Roll) Ahlstrom & Tiffany		*		
Tetrastrum sp.			*	
Unidentified Chlorococcales			**	
Streptophyta				
Closterium acutum Brébisson			*	
Closterium baillyanum (Brébisson ex Ralfs) Brébisson	*	*		
Cosmarium depressum Bailey		*	**	
Cosmarium pseudoholmii O.Borge		*		
Cosmarium spp.	**	*	**	
Elakatothrix gelatinosa Wille		*	**	
Elakatothrix genevensis (Reverdin) Hindák		*		
<i>Elakatothrix lacustris</i> Korshikov			**	
<i>Elakatothrix spirochroma</i> (Reverdin) Hindák		*		
Gonatozygon pilosum Wolle	*	*		
Mougeotia sp.	*			
Spirogyra sp	*			
Staurastrum aniculatum Bréhisson	*			
Staurastrum gracile Ralfs ex Ralfs	**	*	*	
Staurastrum pingue Teiling		*		
Staurastrum planctonicum Teiling		*	*	
Staurastrum sp			**	
Stauratsrum dickiei Ralfs	*			
Staurodesmus dejectus (Brébisson) Teiling			**	
Staurodesmus sp			*	
Teilingia granulata (I Roy & Bisset) Bourrelly			*	
Fuglenonhyta				
Euglena granulata (G A Klebs) E Schmitz		*		
Fuglena polymorpha f minor T Hortobágyi		*		
Fuglena sp			*	
Lenocinclis acus (O F Müller) B Marin & Melkonian				
Lenocinclis acus (Schmarda) B.Marin & Melkonian		*		
Phacus Iongicauda (Ebrenberg) Dujardin		*		
Phacus orbicularis Hühner		*		
Phacus sp			*	
Strombomonas sp		*		
Trachelomonas cf. niora			*	
Trachelomonas hispida (Perty) E Stein	*	*		
Trachelomonas nanctonica Svirenko			**	
Trachelomonas sp			*	
Trachalomonas volvocina (Ebrenberg) Ebrenberg	*		**	
Purrhonhuta				
Caratium cormutum (Ehrenberg) Clanarède & LL achmann		*		
Caratium linum dinalla (O E Müller) Dujardin	*		**	
Ceratium nirunainetta (O.F.Wunter) Dujatum				

Taxon	Periods			
	1972-1975	2010-2012	2016-2022	
<i>Glenodinium</i> sp.		*		
<i>Gymnodinium</i> sp.			**	
Gymnodinium uberrimum (G.J.Allman) Kofoid & Swezy		**	**	
Gyrodinium helveticum (Penard) Y.Takano & T.Horiguchi		*		
Peridinium spp.	**			
Cryptophyta				
Cryptomonas caudata Massart		*		
Cryptomonas erosa Ehrenberg		*		
Cryptomonas ovata Ehrenberg		*		
Cryptomonas spp.	*		*	
Ochrophyta				
Chrysophyceae				
Chromullina sp.			*	
Dinobryon divergens O.E.Imhof	*	*		
Synurophyceae				
Mallomonas acaroides Zacharias			**	
Mallomonas akrokomos Ruttner		*		
Mallomonas caudata Iwanoff [Ivanov]		*		
Mallomonas cf. tonsurata			*	
Mallomonas elongata Reverdin		*		
Mallomonas spp.	*		**	
Synura uvella Ehrenberg	*			
Bacillariophyceae				
Achnanthes sp.		*		
Amphora ovalis (Kützing) Kützing		*		
Amphora sp.			*	
Asterionella formosa Hassall	**	**	**	
Asterionella gracillima (Hantzsch) Heiberg	**			
Asterionella ralfsii W.Smith		*		
Aulacoseira ambigua (Grunow) Simonsen		*		
Aulacoseira granulata (Ehrenberg) Simonsen	*	**	*	
Aulacoseira granulata var. angustissima (O.Müller) Simonsen	**			
Aulacoseira islandica (O.Müller) Simonsen		*		
Aulacoseira italica (Ehrenberg) Simonsen		*		
Bacillaria sp.		*		
Caloneis amphisbaena (Bory) Cleve		*		
Caloneis silicula (Ehrenberg) Cleve		*		
Cocconeis pediculus Ehrenberg			*	
Cocconeis placentula Ehrenberg		*	*	
Cocconeis spp.	*	*	**	
Ctenophora pulchella (Ralfs ex Kützing) D.M.Williams & Round		*		
Cvclotella spp.	*		**	
<i>Cymatopleura solea</i> (Brébisson) W.Smith	*	*		
Cymbella cistula (Ehrenberg) O.Kirchner		*		
Cymbella cymbiformis C.Agardh		*	*	
Cymbella spp.	*			

	Periods		
laxon		2010-2012	2016-2022
Cymbella vertricosa (C.Agardh) C.Agardh		*	
Diatoma ehrenbergii Kützing		*	
Diatoma hyemalis (Roth) Heiberg		*	*
Diatoma sp.			*
Diatoma vulgaris Bory	*	*	
Diploneis ovalis (Hilse) Cleve		*	
<i>Epithemia</i> sp.		*	
Eunotia bilunaris (Ehrenberg) Schaarschmidt		*	
Eunotia minor (Kützing) Grunow		*	
Eunotia soleirolii (Kützing) Rabenhorst		*	
Fragilaria capucina Desmazières	*	*	
Fragilaria crotonensis Kitton	**	**	**
Fragilaria radians (Kützing) D.M.Williams & Round	**		
Fragilaria rumpens (Kützing) G.W.F.Carlson		*	
Frustulia rhomboides (Ehrenberg) De Toni		*	
Gomphonema acuminatum Ehrenberg		*	*
Gomphonema acuminatum var. coronatum (Ehrenberg) Rabenhorst		*	
Gomphonema constrictum Ehrenberg	*	*	
Gomphonema lagerheimii A.Cleve		*	
Gomphonema truncatum Ehrenberg		*	
Gyrosigma acuminatum (Kützing) Rabenhorst		*	
Hannaea arcus (Ehrenberg) R.M.Patrick	*	*	
Melosira varians C.Agardh	*	*	*
Meridion circulare (Greville) C.Agardh		*	
Navicula lanceolata Ehrenberg		*	
Navicula radiosa Kützing		*	
Navicula reinhardtii (Grunow) Grunow		*	
Navicula rhynchocephala Kützing		*	
Navicula spp.	*		*
Navicula vulpina Kützing		*	
Nitschia sp.		*	*
Pinnularia borealis Ehrenberg		*	
Pinnularia spp.	*		
Rhopalodia gibba (Ehrenberg) O.Müller		*	
Stephanocyclus meneghinianus (Kützing) Kulikovskiy, Genkal & Kociolek		**	
Stephanodiscus astraea (Kützing) Grunow		**	**
Stephanodiscus hantzschii Grunow		*	**
Stephanodiscus sp.			**
Surirella sp.		*	
Tabellaria fenestrata (Lyngbye) Kützing		**	**
Tabellaria fenestrata var. asterionelloides Grunow	*	*	
Tabellaria fenestrata var. intermedia Grunow	*		
Tabellaria flocculosa (Roth) Kützing	*	**	**
Ulnaria acus (Kützing) Aboal	**	*	
Ulnaria biceps (Kützing) Compère		*	
Ulnaria ulna (Nitzsch) Compère	*	*	*

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