ROLE OF THE PLANKTONIC COMMUNITIES IN THE REGULATION AND INDICATION OF EUTROFICATION PROCESSES IN SHALLOW MOUNTAIN WETLANDS

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Protection and restoration of wetlands requires correct evaluation of their ecological and trophic state, i.e. detection and verification of appropriate indicators. The aim of the study is to clarify the role of planktonic communities (phytoplankton and bacterioplankton) in eutrophication processes in shallow landslide-dammed lakes and to apply routine indicators of eutrophication processes for a case study of Dragichevo lake. From May 2001 through September 2002 a bathometric map and map of macrophyte distributions was constructed, changes in the main hydrochemical parameters, phyto- and bacterioplankton were tracked, and the Carson's trophic state index determined. The results suggested that re-suspension processes and active interaction between sediments and water significantly influence both the plankton communities and chemical parameters of these wetlands.

1. Introduction

The protection and restoration of headwater wetlands requires the correct evaluation of their ecological and trophic state, which is connected to the detection and verification of appropriate biological and chemical indicators. Most investigations of wetlands emphasise the quality and

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composition of macrophyte communities and their relation to the water quality. Many investigations address the processes in the sediments and in the root zone of the macrophytes, especially in the context of natural wastewater treatment systems. However, there is also increasing interest in the planktonic communities of wetlands and their role in eutrophication processes. The aim of this study is to clarify the role of planktonic communities (phytoplankton and bacterioplankton) in eutrophication processes in landslide lakes using a broad range of routine indicators for estimation of trophic state.

2. Materials and Methods

2.1. SAMPLE COLLECTION

This study was carried out from May 2001 to September 2002. Water samples were collected at 0,2 m below the water surface from the pelagic zone of the lake (Kiselev,1956).

2.2. ABIOTIC PARAMETERS

Water temperature and oxygen content were measured with oxymeter - "OXI-196" and pH with "pH 323/set" field equipment manufactured by Wissenschaftlich-Technische Werkstetten (WTW). The chemical oxygen demand (COD) was measured by "WTW-mini test"(ISO 14 000). The biochemical oxygen demand (BOD1, BOD3, BOD5) was measured according to APHA (1982) (without sample pre-treatment). Total suspended solids (TSS) was determined by the "dry weight technique" (APHA, 1982) and chlorophyll-a (chl-a) was measured spectrophotometrically (ISO10260).

Ammonia (NH4-N), nitrite (NO2-N), nitrate (NO3-N) and phosphate (PO4-P) were determined spectrophotometrically with laboratory kits (MERCK). The codes of the methods are: NH4-N method # 14752; NO3-N method # 14773; NO2-N method # 14776; PO4-P method # 14848 and total phosphorus (TP) - method # 14848 after digestion with per-sulfate and sulphuric acid at 120oC for two hours. Alkalinity was determined according to ISO 9963-1 by titration with 0.1 N HCI and brom-krezol green – methyl rot as an indicator.

2.3 BIOTIC PARAMETERS

Water samples for phytoplankton (0,5l) were fixed with lugol solution then gravity concentrated to 30ml final volume of the settled samples. The enumeration of the phytoplankton was carried in Burker chamber and the results presented in cells per cubic meter. The species were determined and enumerated with microscope (Amplival) at magnification x400. Total bacterial count (TBC) was determined by direct microscopic enumeration on membrane filter after staining with erythrosine [9]. Saprophytic bacteria (SB) were enumerated by indirect "plate count" technique according to the routine microbiological practices [8]. Saprophytic bacteria were cultivated on organic-enriched agar. Trophic indices were calculated by the method of Carlson [3].

3. Site description

The system of landslide lakes known as the Dragichevo wetlands is situated on the southern slopes of Ljulin Mountain between 870 and 950m.a.s.l. near Dragichevo village and 22 km away of Sofia. Dragichevo Lake is the biggest (Table 1) of thirteen wetlands, most of which dry up in the summer.

Parameter	Value
Maximum length	95m
Maximum width	38m
Maximum depth	3,1m
Mean depth	1.11m
Surface area	3 035m^2
Volume	3 045m^3

Table 1. Morphometrical parameters of Dragichevo lake.

This lake is divided into two sub basins with different morphometry: a northern with maximum depth of 3,1m and a southern with depth of 1,1m (Figure 1). The bottom sediments of the lake are also distinctly different in the sub basins. The shallower part of the lake is covered with a thick layer of organic sediments while, in the deeper part, the sediments are more dense and lack macrophyte debris.

Apart from the western bank and a small part of the eastern bank, the lake is surrounded by a belt of emergent vegetation – *Typha latifolia*. Approximately 80% of the lake surface is covered by dense submerged vegetation – *Potamogeton natans* mixed with two species of *Chara* and around the bank *Callitriche cophocarpa* with possibly some *Sparganium minimum*. The sub basins are divided by shallow waters, and at low water level a mud bank almost separates the basins. A patch of *Schoenoplektus lacustris* has recently started to overgrow the *Potamogeton* at that place (Figure 2), which is evidence of the rapid loss of depth.

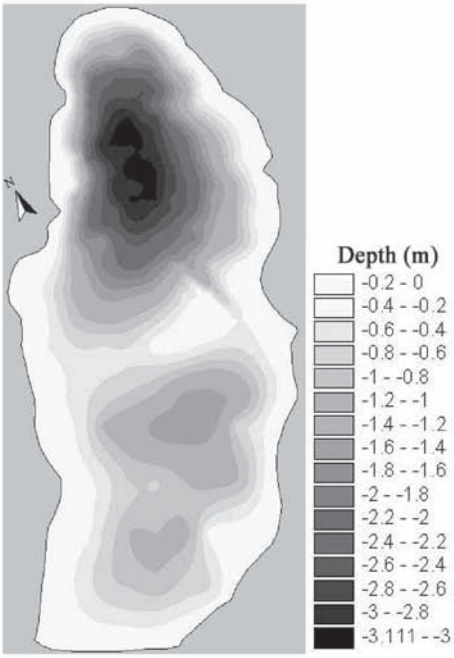

Figure 1.Bathomeric map of Dragichevo lake.

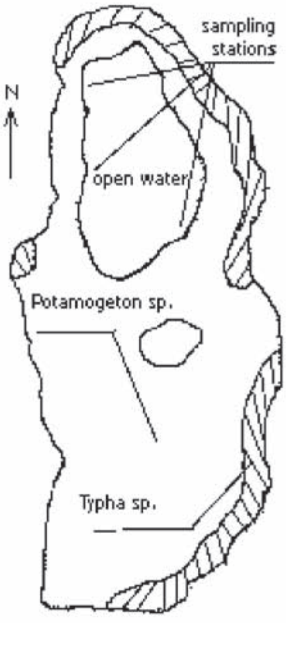

Figure 2. Schematic vegetation map of the Dragichevo lake.

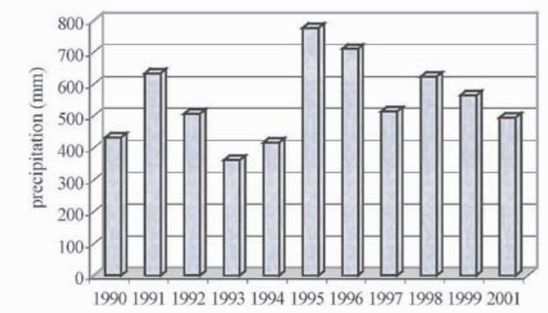

Figure 3. Annual precipitation.

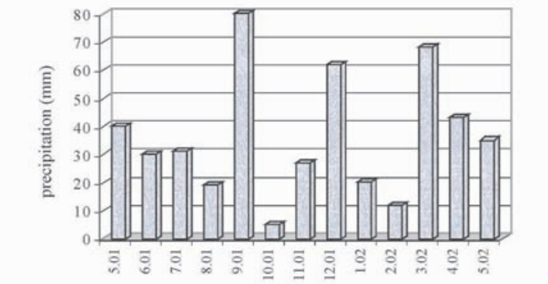

Figure 4. Monthly precipitation.

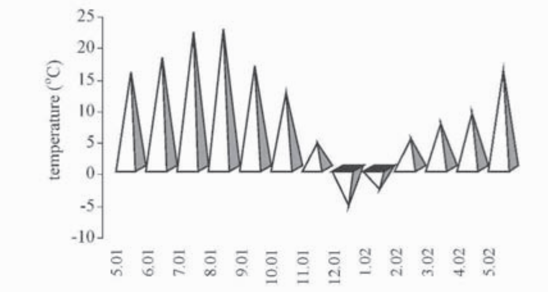

Figure 5. Mean-monthly air temperatures.

The landslide wetland system has small catchment - cca 100 ha, 70 ha of which are above Dragichevo Lake. The upper catchment is covered by cultivated forests of *Pinus nigra, Populus deltoides, Populus tremula* and remains of a natural forest of *Quercus pubescens*. The water level in the wetlands is regulated by surface runoff and groundwater supplies. Mean monthly (Figure 3) and annual (Figure 4) precipitation vary strongly. The annual amplitudes of air temperatures are high (Figure 5). This determines the large changes in the temperature and chemical parameters of the lake.

In periods of high precipitation and snow melt the ponds in the wetland system are connected by a temporary brook that transforms into a small river at the lower end of the cachment, then flows down to join the Strouma River.

In the past, drainage channels had been dug at the lower part of each wetland decreasing the water level in the individual wetlands by 30 to 50 cm to the detriment of the whole system. Due to the decreased water level, the area of the open water in the system has shrunk and most, apart from Dragichevo Lake, has been overgrown by macrophytes. This drying up threatens the existence of rare and endangered species as: *Chara, Emys orbicularis, Misgurnus fossilis, Salamandra salamandra, Triturus vulgaris, Hyla arborea.*

4. Chemical variables

4.1 TEMPERATURE

The average value and amplitude of the water temperature in 2001 was lower than the average water temperature for the corresponding period of 2002 (Figure 6). The high range of water temperatures in Dragichevo Lake is due to the strong water level fluctuations in the wetland. The lower water temperatures in 2001 are probably the cause of the low phytoplankton numbers in 2001, as well as the higher concentration of oxygen relative to 2002.

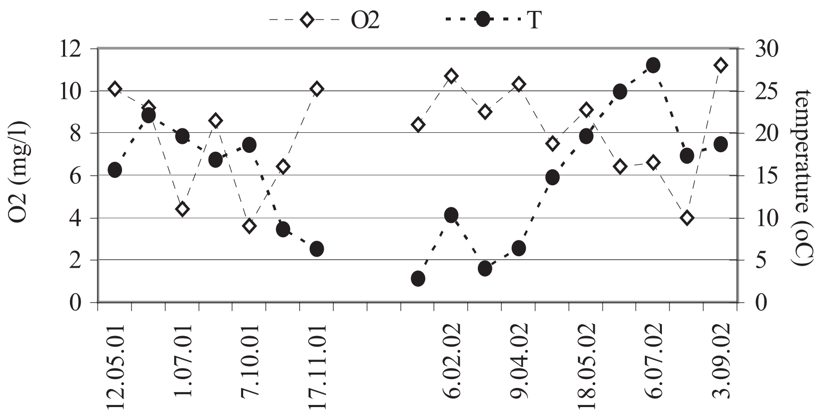

Figure 6. Annual changes of oxygen concentration and temperature in Dragichevo Lake.

4.2 OXYGEN

Comparing the May to September periods of the two years, the oxygen concentration in 2001 was 0,6 mg/l higher than the average for 2002 and the oxygen saturation greater by 10%..The annual amplitude of variation of oxygen content in Dragichevo Lake was high. The period with oxygen deficit was twice as long as the period of oxygen over- saturation, which indicates intensive respiration and biodegradation. So, changes in oxygen content were affected by the high amplitude of water temperature change, changes in the water level and variations in phytoplankton development.

4.3 NUTRIENTS

The annual changes of NH_4-N, NO_2-N and NO_3-N are presented on Figure 7. The concentrations of ammonium and nitrate nitrogen in the lake are almost twice as high (and those of phosphorus twice as low) as data from similar Bulgarian lakes [2]. The maximum values of NH_4-N measured in the autumn were 1,5 to 3 times higher than the average and in the summer of 2002 – 3 times the average. The peaks in NH_4-N concentration coincide with the periods of rapid decrease in oxygen concentration and increases in phytoplankton numbers. Minimum concentrations were measured in the spring of 2002.

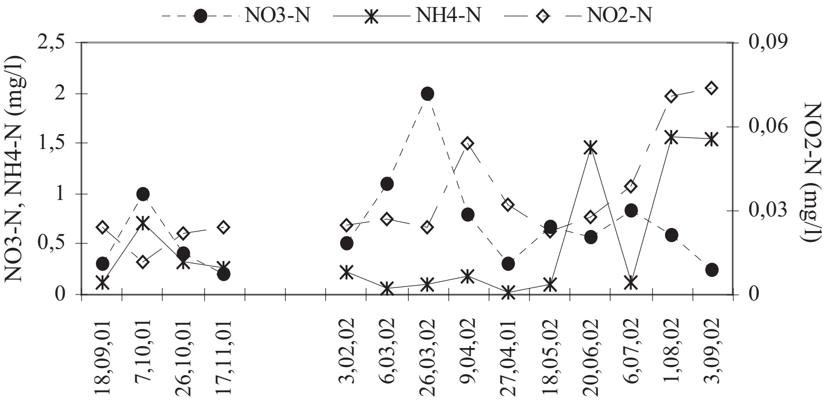

Figure 7. Annual changes of NH_4-N, NO_2-N and NO_3-N.

Maximum concentrations of NO_2-N coincide with the peaks of NH_4-N. In August and September 2003, concentrations of NO_2-N were twice the average. In the autumn of 2001, a peak of NO_3-N (1mg/l) as well as another one in March 2002 (1,1-2 mg/l) was observed (Figure 7).

The periods of low PO_4-P correspond with high values of chlorophyll-a concentration (5-254 ug/l) (Figure 8). It is possible that plankton rapidly recycle nutrients and thus sustain their development under low PO_4-P concentrations [16] or possibly, significant amounts of phosphorus in the lake weres incorporated into the biomass of the phytoplankton and/or bounded in metal complexes in the sediments [4,5,6].

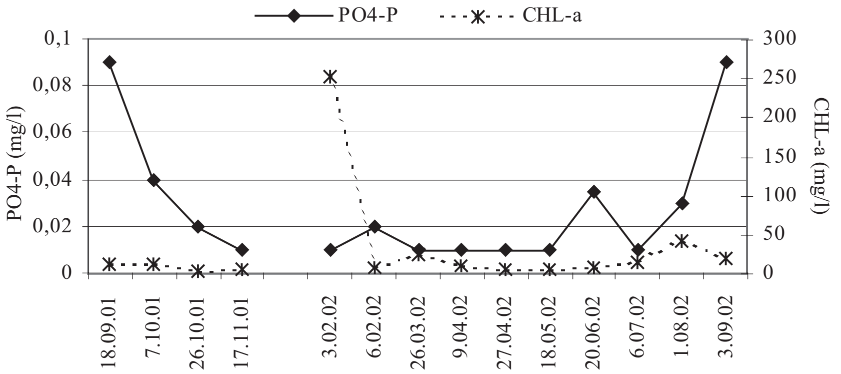

Figure 8. Dynamics of chl-a and PO4-P in Dragichevo Lake.

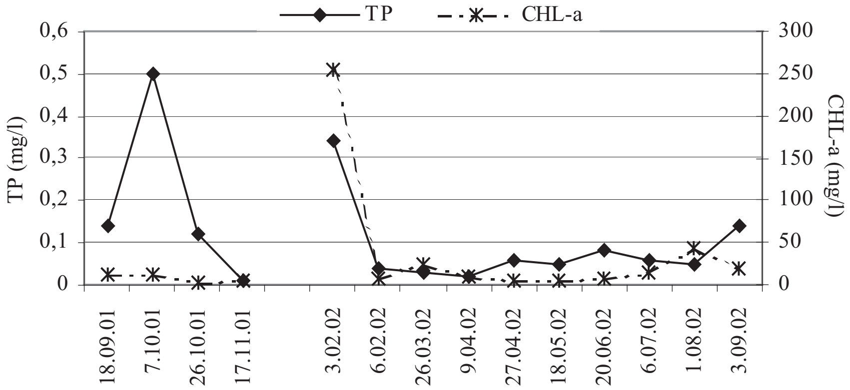

Figure 9. Dynamics of chl-a and TP in Dragichevo Lake.

Changes of TP and PO_4-P are similar - two peaks in the autumn periods and increased concentrations in July 2002. The highest value of TP was measured in October (0,5 mg/l). The autumn of 2001 is characterized also by high levels of total suspended solids (average 13,5 mg/l). Strong increases in TP were measured in February 2002 during the winter bloom of the phytoplankton. For the most part TP concentrations remained in the range 0,1-0,8 mg/l (Figure 9). By contrast, TSS data have complex dynamics and show additional peak in April 2002 (Figure 10).

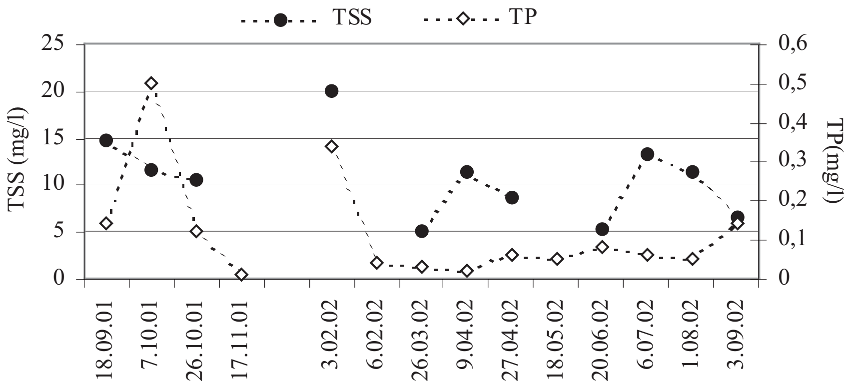

Figure 10. Changes in total suspended solids and total phosphorus concentrations throughout the investigated period.

4.4 pH

The pH of the lake waters is neutral to slightly alkaline and remained. almost constant (6,8 - 7,5) throughout the study period with only one exception in May 2002 (pH = 5). This was due, probably, to the high buffer capacity of the water (alkalinity of 2,8-4,0 mg/eqv).

4.5 BIOCHEMICAL AND CHEMICAL OXYGEN DEMANDS (BOD AND COD)

Changes in organic substances in the lake were studied through measurements of COD and BOD (BOD1, BOD3, and BOD 5. The values of COD were high throughout the investigation (average 200mg/l) and did not correlate with other measured parameters.

BOD5, BOD3, and BOD1 had parallel patterns of variation (Figure 11). Unusually, oxygen and BODx also changed in parallel (Figure 12). A correlation between BODx and chlorophyll-a concentration was also observed. Evidently, the phytoplankton possesses a strong influence on the values of BODx [14, 15], while COD was influenced by humic substances, which is typical for shallow lakes [16].

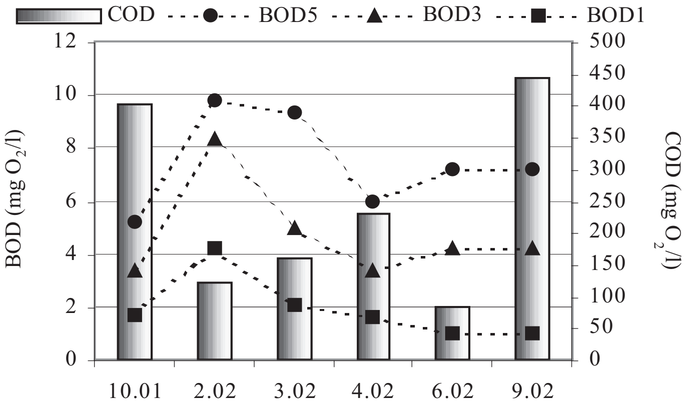

Figure 11. Dynamic of the COD and BOD1, BOD 2, BOD 3 in the lake.

5. Biological parameters

5.1 PHYTOPLANKTON

The Dragichevo Lake phytoplankton includes 83 species of 9 groups. Despite the high diversity of species (Table 2) and groups (Figure 13) compared to other Bulgarian wetlands [11], the group diversity of the phytoplankton is generally low. The highest diversity of groups is observed in July 2001 and September 2002.

Group		Number of species
Cyanophyta		7
Zygnemophyta		10
Cryptophyta		2
Bacillariophyta		18
Chlorophyta		24
Xantophyta		2
Euglenophyta		14
Dinophyta		6
Chrysophyta		2
Total	9	83

Table 2. List of phytoplankton species in Dragichevo Lake.

Cryptophyta were dominant in the lake from summer 2001 to April 2002 with two exceptions only, when Bacylariophyta were the most abundant.

In May 2001 Chlorophyta prevailed followed by Dinophyta, while in the same period of 2002 successive blooms of Dinophyta (May) and Chlorophyta (June) were observed.

There were extremely low quantities of Cyanophyta in 2001, while in 2002 two peaks in their numbers were observed. The Cyanophyta reached 24% in April and 21% in July mostly due to *Anabaena sp*. Presence of *Microcystis sp.* was not encountered throughout the investigated period. The success of the Cyanophyta is strongly dependent on the humic compounds in the waters and the circulation pattern of the system [10].

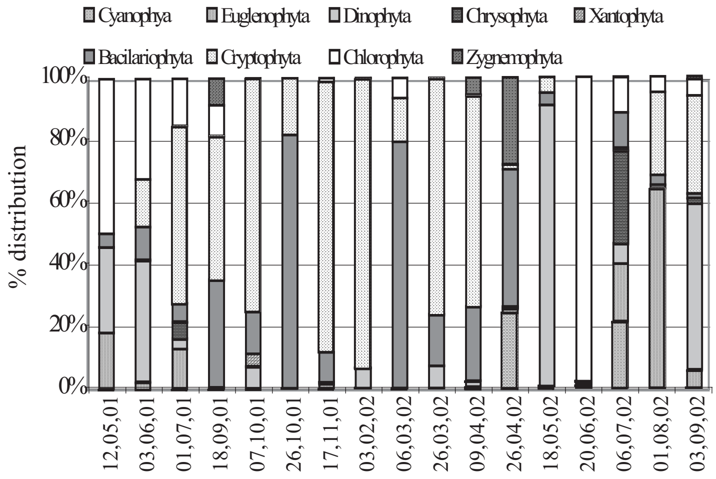

Figure 12. Distribution of the different phytoplankton groups in the samples.

Such seasonal variability in the different phytoplankton groups is typical for polymictic eutrophic lakes. The pattern is similar in both years but higher temperatures in 2002 allow the appearance of the prevailing groups one month earlier.

The numbers of the phytoplankton in the lake waters varied between 16,3x106 to 623x106 cells/m3 (Figure 13) with an average of 133,3x106 cells/m3. The number of phytoplankton in 2002 was 5 times higher than in 2001. The maximum was in June of both years.

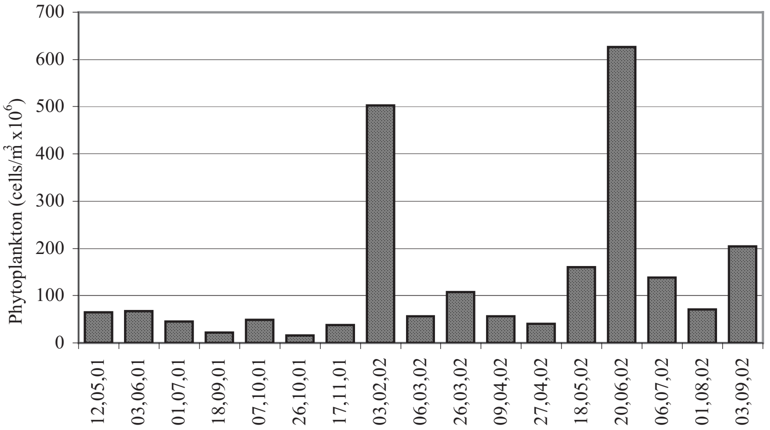

Figure 13. Changes of the phytoplankton cell numbers in the samples.

Differing from the phytoplankton numbers, the chlorophyll-a (chl-a) concentration in the second half of 2001 is slightly higher compared to the same period of 2002 (Figure 14). The average chl-a concentration in 2001 was 24 ug/l, while in 2002 it was 18 ug/l. The maximum measured chl-a concentration – 254 ug/l was during the winter bloom of Cryptomonas sp. in February 2002 and the minimum - 2.6 ug/l was measured in October 2001. The minimum and average chl-a concentrations for the whole 2002 are 5 ug/l and 39 ug/l, respectively.

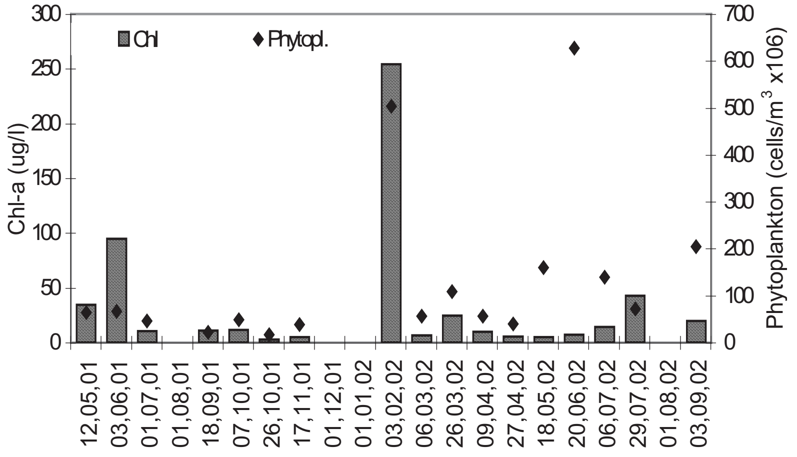

Figure 14. Quantity of chlorophyll-a and phytoplankton in Dragichevo lake.

Throughout the investigated period, chl-a concentration correlates significantly with numbers of phytoplankton (r2 = 0.89) (Figure 15). Such dependence was not observed from May to September 2002 (r2 = 0.27). In the summer of 2002, mass development of Dynophyta (*Dinobrion sp.* – 90%) and Chlorophyta (*Monoraphidium contortum* – 98%) led to the biggest discrepancies in the relationship.

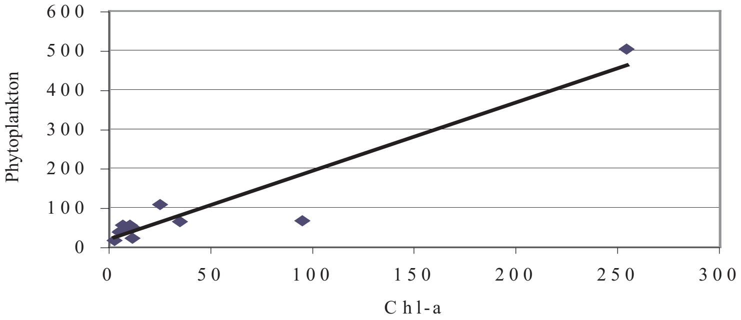

Figure 15. Correlation between phytoplankton and Chl-a, May 2001 – April 2002.

5.2 BACTERIOPLANKTON

In contrast to phytoplankton, the quantity of bacteria was low and relatively stable. The total bacterial count (TBC) varied between 40 – 200 x 103cells/ml, excepting a spike in October 2001 (1008x103 cells/ml) (Figure 16) when peaks in TP and COD were measured too. The quantity of saprophytic bacteria (SB) was also low and stable (1-4x103 cells/ml).

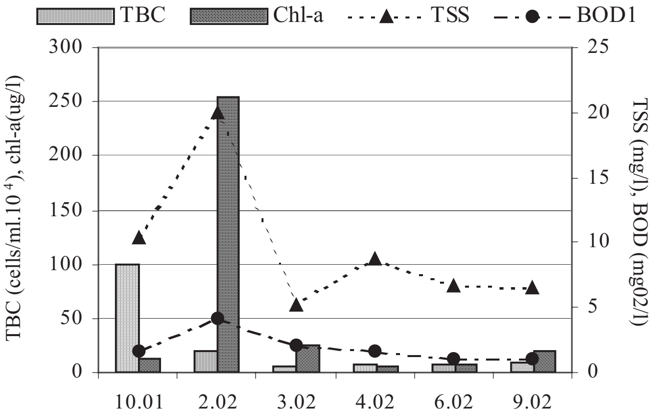

Figure 16. Changes in total bacterial count in relation to chlorophyll-a, total suspended solids and BOD1.

TBC showed relationship only with the suspended solids in the water (rs = 0,714) (Asenova et al., 2003) and did not depend on the concentration of organic substances measured as BODx and COD. Therefore, the quantity of bacteria was influenced more by resuspension than by the amount of available organic substances.

5.3 TROPHIC STATE INDICES

The average values of the Carlson's trophic state index derived on the basis of chl-a concentration (TSICHL) and total phosphorus (TSITP) are 56,8 and 64,2, respectively (Figure 17). These data suggest possible phosphorus limitation for the following months; November 2001, March – April and August 2002. Otherwise, the higher values of the TSITP could indicate that factors other than phosphorus limit the development of phytoplankton. In October 2001 the values of the TSITP are almost twice those of TSICHL, again due to the re-suspension of bottom sediments following a series of storm events with heavy rainfall. Increase in TSITP is also observed after the spring rains in 2002. The winter maximum of TSITP was entirely due to phytoplankon-incorporated phosphorus.

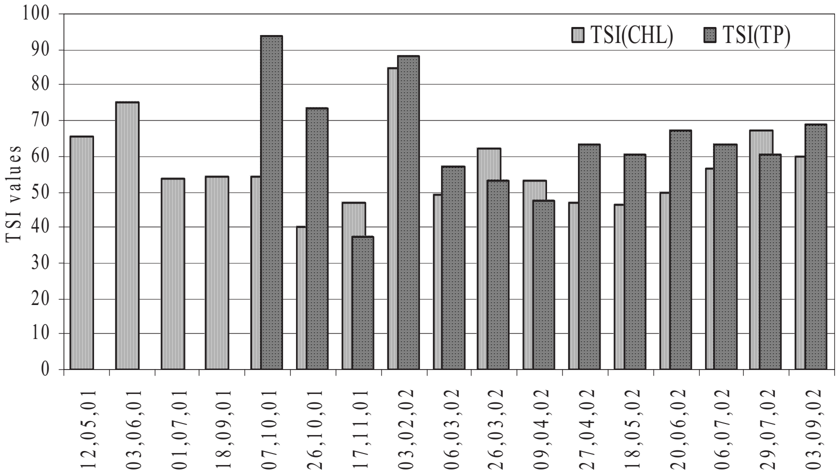

Figure 17. Carlson's trophic state indices calculated on the basis of chlorophyll-a concentration (TSICHL) and the amount of total phosphorus (TSITP).

6. Discussion

6.1 CHEMICAL PARAMETERS

The highly pronounced annual amplitude of the water temperature variations in Dragichevo Lake is due to strong water level fluctuations. These are determined by environmental factors and human activities in the catchment. Despite the polymictic nature of the lake, the periods with oxygen deficit prevailed over the periods with oversaturation, which indicates intensive respiration and biodegradation processes in the wetland.

The twice higher concentrations of ammonium and nitrate nitrogen in the lake are mostly due to the input of nitrogen as faecal material from the animals in the surroundings of the lake and from active decomposition. The low phosphorus concentrations, compared to data from similar Bulgarian lakes [2], are due to the absence of human settlements in the catchment. In the periods of ice cover during the winter peak of the phytoplankton, concentrations of NO_3-N, NO_2-N, NH_4-N and PO_4-P were below average because of their uptake in the phytoplankton. After the winter maximum of phytoplankton, the concentration of NO_3-N increases and those of NH_4-N remain low, indicating active nitrification during the spring. In the summer and autumn, concentrations of NO_3-N decreased. The opposite tendency was observed in NO_2-N. This was due to incomplete nitrification in the summer and/or active denitrification. Denitrification processes predominate in dense vegetation during late summer and early autumn. Uptake from the water column is active in the early summer during the rapid growth of the macrophytes.

During the March peak in NO_3-N concentration mass development of Dinophyta was observed. The summer peak of NH_4-N coincided with a peak in Chlorophyta. The autumn increase in NO_2-N concentrations corresponded with high levels of Zygnemophyta and Cryptophyta in the plankton. Of course, the nutrients influence the amount of the phytoplankton and vice versa.

The changes in the TP concentrations are similar to those of chl-a in 2002, while such connection was not observed in the autumn of 2001. The discrepancies in 2001 were due to heavy rains, which increase both TP and PO_4-P concentrations in the waters. In the absence of external forcing, the amount of TP in the water is mostly formed by phytoplankton. A substantial part of the readily available phosphorus in shallow lakes is found in sediments rather than in the water column. Therefore, the traditionally used "total-P" is far from being a perfect indicator for the nutrient status of shallow lakes. In shallow lakes, a considerable part of the available phosphorus is stored in the sediments and rapidly returns to the water column during resuspension [13].

Although, the free water surface in the lake is small compared to the macrophyte beds, the phytoplankton plays a significant role in the dynamics of nutrients in the water. It tends to increase the total concentration of nutrients in the water column, especially the concentration of phosphorus by two main mechanisms: high photosynthetic activity causes the pH in the water to rise and in this conditions the capacity of iron to bind phosphorus decreases; phytoplankton remove PO_4-P from the water column and stimulate the desorption of phosphorus from sediment and suspended particles. In the case of Dragichevo Lake, the second mechanism most probably prevails as the water in the lake is well buffered, pH is relatively stable, and the sediments except iron, contain also manganese and other metals in high concentrations [12].

The COD and BODx data show that considerable concentrations of organic substance exist in the wetland. COD and BODx did not correlate with parameters such as: TBC, quantity of saprophytic bacteria, and TSS. Even more, there is no correlation between both parameters for the investigated period, which should be present if we consider the fact that easily degradable organic substances increase the proportion of BOD in COD.

The high values of COD and the absence of correlation with all other parameters are linked to high amounts of humic substances in the water. BOD correlates with chl-a only and follows the changes in the oxygen concentration. Here, the BOD was influenced by the phytoplankton. It can be confirmed that in the Dragichevo Lake, COD and BODx do not reflect, correctly, the amount of autochthonous organic load in the wetland.

6.2 BIOLOGICAL PARAMETERS

Highly pronounced peaks of different phytoplankton groups with high deviations from the mean value, typical for shallow polymictic lakes, were observed in the Dragichevo Lake. Microcystis sp. was not found and there were no blooms of Cyanophyta in the lake. This supports the suggestion that the lake is not nitrogen limited, which in combination with the diverse environment and external forcing decreases the possibility of Cyanophyta blooms.

The high correlation between the quantity of the phytoplankton and chl-a concentration gives the possibility to use the relationship for estimation of the phytoplankton quantity, which would make the monitoring much easier and faster. In the wetland this correlation was disturbed by the development of certain phytoplankton species like Dinobrion sp. and Monoraphidium sp. In such systems the use of total chlorophyll concentrations as a monitoring tool could be more appropriate.

The quantity of bacteria in Dragichevo Lake is low for eutrophic water body and this parameter has significant positive correlation with TSS only. Positive correlation was observed between the growth rate of bacteria and organic substances too [1]. Therefor, the amount of organic substances influenced much more bacterial activity than bacterial count. Important limiting factor of the bacterial quantity is predation of zooplankton and reduction of resuspension by submersed macrophytes, but those relationships have not been studied in the lake yet.

The observed discrepancies between the trophic indices (TSITP and TSICHL) were entirely determined by external forcing, such as rain events and wind. The values of the indices in calm weather are close to the theoretical equilibrium and suggest that most of the total phosphorus is incorporated into the biomass of the phytoplankton. The TSICHL index is more suitable for determining the trophic state in small wetlands, as it reflects the actual amount of the phytoplankton and is not so strongly dependent from sporadic events in the catchment.

7. Conclusion

In a detailed study of a shallow landslide-dammed, the accent was put on the characteristics and role of planktonic communities. The results suggested that re-suspension processes (connected to rain events and wind action) influenced both the plankton communities and dynamics of chemical parameters significantly.

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