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Limnological research of acidified lakes in Czech part of the Šumava Mountains: trophic status and dominance of microbial food webs

Limnologický výzkum acidifikovaných jezer na české straně Šumavy: trofický status a dominance mikrobiálních potravních řetězců

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Abstract

Chemical and microbiological status of glacial lakes in the Czech part of the Šumava Mountains (Central Europe) was studied from 1986 to 1994. All lakes were strongly acidified, with sulphate and nitrate the dominant anions, and elevated aluminium concentrations. According to their trophic status, the lakes were divided into two groups: oligotrophic (Černé L., Čertovo L.) and mesotrophic (Prášilské L., Plešné L.) lakes. Nitrate was the major form of total nitrogen (TN) in all lakes. Concentrations of total phosphorus (TP) and dissolved organic carbon were low in the oligotrophic lakes (~ 4 μg l^{-1} and ~ 1.5 mg l^{-1}, respectively), while nearly twice as high in the mesotrophic lakes. Anoxia in the bottom part of the hypolimnion affected concentrations and forms of nutrients in the mesotrophic lakes during summer and winter stratification. High TN to TP and particulate C to particulate P ratios, as well as high phosphatase activities during the elevated concentrations of chlorophyll *a,* indicated a phosphorus limitation of phytoplankton in all lakes. The ecologically unique type of aquatic ecosystem has developed in these lakes where higher trophic levels (zooplankton, fish) disappeared due to acidification and the microbial loop became dominating the pelagic food webs. Low primary production and allochtonous input of organic matter seem to alternate as the carbon and energy sources for the active bacterioplankton controlled by both heterotrophic and mixotrophic flagellates. Only in Prášilské Lake where crustacean zooplankton persisted (likely due to a lower concentration of total aluminium), also heterotrophic microbes were an additional food source for daphnid population besides phytoplankton.

Key words: mountain lakes, acidification, nutrients, ectoenzymes, plankton, Bacteria, Protozoa, microbial loop

Introduction

Fresh waters of large European and American regions, characterized by bedrocks that are resistant to chemical weathering, have been affected by anthropogenic acidification (e.g. LIKENS & BORMANN 1974, WRIGHT & HENRIKSEN 1978, PSENNER & CATALAN 1994). The acidification has become a dominant factor disturbing and even controlling the quality of water and biota of the lake ecosystem (HENRIKSEN 1980, CHARLES & al. 1989). The results of whole lake acidification experiments of SCHINDLER & al. (1985) and BREZONIK & al. (1993) were consistent with changes in phytoplankton and zooplankton species, which were reported from synoptic surveys of lakes in regions of high acid deposition (SCHINDLER 1988, 1994). Similar acidification-derived changes have been also reported for lakes in former Czechoslovakia, both in the High Tatra Mts. and Šumava Mts. (STUCHLÍK & al. 1985, FOTT & al. 1987, 1992, 1994, LUKAVSKÝ 1994).

Human impact on water and biota composition has been well documented for the lakes in the Šumava Mts. (Bohemian Forest), owing to a more than one-hundred-year tradition of limnological research there. The acidification of the lakes, accompanied with a drop of pH and an increase of nitrate concentration, began in the 1960s (FOTT & al. 1987, VESELÝ & MAJER 1992). Nowadays the lakes are fishless and most of metazooplankton disappeared in all but one lake (FOTT & al. 1994, VESELÝ 1994). The pH-related changes in the composition of zooplankton and diatom species and deposition rates of heavy metals and carbonaceous particles have been well documented in lake sediments (SCHMIDT & al. 1993, VESELÝ & al. 1993, PRAŽÁKOVÁ & FOTT 1994). During the last two decades, an extensive hydrobiological research has recorded a recent acidification status and particular changes in water chemistry of these lakes (VESELÝ & MAJER 1992, FOTT & al. 1994, KOPÁČEK & al. 1995). However, no consistent results have been published on microbiology of these lakes so far.

The aim of this study is to present data on the nutrient and microbiological status of lakes situated in the Czech part of the Šumava Mountains and elucidate possible pelagic food webs in some of these lakes.

Site description

In the Šumava Mountains (13° to 14° E, 49° N; maximum altitude 1456 m a.s.l.) there are 8 lakes of glacial origin. Five lakes are in the Czech Republic. Their basins are small, with steep slopes covered predominantly by spruce. The bedrock is of biotite-rich paragneiss, together with gneiss, quartzite and granite. Most of the lake areas have been affected by various human activities (e.g., iron mining, glass works, pasturing, forestry) since a half of the 17th century (for review, see VESELÝ 1994). Nevertheless, most kinds of land use have been reduced in the lake basins after 1960 when the Šumava Mts. became a protected landscape area. The Czech as well as German names and main physical characteristics of the lakes are shown in Table 1.

Average rates of wet-only atmospheric deposition of NH_4-N, NO_3-N and SO_4-S over the Czech Republic in the midle 1980s were 8.3, 6.1 and 21 kg $ha^{-1} yr^{-1}$, respectively, as estimated from 12 Czech EMEP stations (MOLDAN 1991). In the 1980s, the Šumava Mts. were ex-

Table 1. Main physical characteristics of lakes in the Czech part of the Šumava Mountains (according to Chábera et al. 1987).

Parametr	Černé[1)]	Čertovo[2)]	Prášilské[3)]	Plešné[4)]	Laka[5)]
Altitude (m a.s.l.)	1008	1028	1079	1090	1096
Lake area (ha)	18.4	10.3	3.7	7.5	2.8
Max. depth (m)	40	36	15	18	4
Mean depth (m)	15.6	17.9	7.4	8.2	1.4
Volume (mil. m^3)	2.88	1.85	0.27	0.62	0.04
Catchment area (ha)	129	86	52	67	135
Catchment area to Volume ($m^2 m^{-3}$)	0.45	0.46	1.93	1.08	33.75

[1-5)] German names of the lakes: [1)] Schwarzsee, [2)] Teufelsee, [3)] Stubenbachsee, [4)] Plöcknesteiner See, [5)] Lakka.

posed to 11–18 kg ha^{-1} yr^{-1} of wet-only N deposition and to about 35 kg ha^{-1} yr^{-1} of total N deposition (VESELÝ & MAJER 1992). Total sulphur deposition was estimated at 30 kg ha^{-1} yr^{-1} in 1990 (VESELÝ & MAJER 1992). During the 1990s, deposition of S and N followed decreasing trend due to a drop in S and N emissions in Central Europe, intensified also by political and economical changes in the Czech Republic in 1989 and relatively mild winters compared to the 1980s (KOPÁČEK, unpubl. data).

Methods

The lakes were sampled irregularly from 1986 to 1994, usually in autumn. From 1991 to 1993, particular attention was paid to Černé L., Plešné L. and Prášilské Lake which were sampled in spring and autumn, the latter also in winter. In 1994, Plešné Lake was sampled 8 times from spring overturn to winter stratification. Samples were taken from the surface (0.1 to 0.6 m depth) and, in Prášilské L. and Plešné Lake, also from 0.5–1.0 m above the bottom of the lake at their deepest point. The samples were immediately filtered through a 100 µm polyamide mesh to remove large particles, stored in darkness at 4°C, and analyzed within 2 days.

NH_4^+-N content was determined by the rubazoic acid method and NO_3^--N spectrophotometrically after reduction to NO_2^--N using alkaline hydrazine. Total organic nitrogen (TON) content was determined by Kjeldahl combustion followed by a colorimetric determination of ammonia using Nessler's reagent. Total nitrogen (TN) was a sum of all of the above amounts. Total phosphorus (TP) was determined by perchloric acid digestion, chemical oxygen demand (COD) by the dichromate semi-micro method, dissolved organic carbon (DOC) and particulate organic carbon (POC) with a Liqui TOC analyser (Foss-Hereaus, Germany). Inductively coupled plasma spectrometry and capillary isotachophoresis technique were used for analyses of metals (Ca, Mg, Na, K and total aluminium, Al_{tot}) and sulphate, respectively. Chloride was determined spectrophotometrically with a Hg thiocyanate method. For more details on the methods see, e.g., KOPÁČEK & STUCHLÍK (1994) and KOPÁČEK & al. (1995).

Phytoplankton biomass was estimated as chlorophyll *a* concetration analyzed fluorometrically after extraction into 90% acetone (VYHNÁLEK & al. 1994). Primary production was measured by the ^{14}C method (RIEMANN & SONDERGAARD 1986); dark fixation was not subtracted. Samples were exposed in 5 depths of the lake (up to double Secchi depth) for two hours. An abundance and biomass of bacteria and heterotrophic nanoflagellates (HNF) were determined in an epifluorescence microscope after DAPI staining (PORTER & FEIG 1980). Bacterial production was measured with incorporation of ^3H-methylthymidine (RIEMANN & SONDERGAARD 1986). Total ectoenzymatic activities were measured fluorometrically using MUF substrates (HOPPE 1993). Besides the total activities, the share of dissolved enzymes was determined in the 0.2 µ filtrate (VRBA 1992). Protistan ingestion rate on bacteria was estimated using fluorescently stained bacteria (SHERR & SHERR 1993).

Results and discussion

Abiotic factors, water chemistry and nutrients

All lakes in the Šumava Mts. are acidified to a different degree. In general, the lakes with the lowest catchment area to volume ratio (Table 1) are the most severely acidified ones and vice versa (Table 2). Concentrations of bicarbonate were always zero in the surface water of the lakes, except for Laka where alkalinity ranged between 5 to 20 µeq l^{-1} throughout the study. Laka Lake was also the only lake with pH above 5, while pH values between 4.10 and

Table 2. Chemical characteristics of the surface samples for the Šumava lakes. Mean values (range in parentheses) are given for the sampling period and number of samples (n) as indicated.

Lake		Černé	Čertovo	Prášilské	Plešné	Laka
Sampling	period n	1986-1994 12	1986-1994 6	1988-1994 11	1990-1994 14	1986-1993 4
NH_4-N	μg/l	24 (5-46)	36 (9-57)	53 (6-130)	50 (10-150)	5 (<5-9)
NO_2-N	μg/l	(<1-17)	(<1-1)	(<1-5)	(<1-19)	(<1-2)
NO_3-N	μg/l	1056 (908-1230)	694 (490-970)	635 (280-1020)	370 (220-720)	724 (520-1130)
TON	μg/l	178 (130-240)	223 (149-320)	312 (210-450)	311 (245-460)	286 (220-350)
TP	μg/l	4.1 (2.2-6.3)	4.2 (2.5-6.4)	5.0 (4.0-7.2)	8.9 (5.6-13.0)	6.8 (4.2-8.3)
COD	mg/l	3.6 (1.8-7.7)	5.3 (4.3-6.6)	8.9 (5.8-15.8)	7.7 (4.5-11.8)	8.7 (6.4-11.8)
DOC	mg/l	1.25 (0.67-1.8)	1.76 (1.42-2.1)	3.62 (2.7-4.5)	2.53 (1.34-4.6)	3.36 (3.0-3.6)
Chl *a**)	μg/l	2.31 (0.30-3.7)	(3.0–3.25)	3.39 (0.4-7.2)	4.90 (1.5-12.1)	(1.2–3.4)
pH		(4.45-4.87)	(4.05-4.50)	(4.60-4.89)	(4.53-4.99)	(4.86-5.80)
SO_4^{2-}	mg/l	6.0 (5.4-7.4)	7.7 (6.7-9.3)	4.3 (3.2-6.5)	7.3 (6.6-8.6)	2.23
Cl^-	μ/l	830 (760-940)	800 (690-940)	690 (630-720)	630 (550-710)	830 (810-840)
Ca^{2+}	μg/l	950 (730-1120)	680 (440-780)	690 (500-870)	1180 (910-1580)	860 (770-920)
Mg^{2+}	μg/l	550 (440-650)	440 (300-520)	380 (270-460)	270 (130-390)	460 (370-500)
Na^+	μg/l	870 (770-960)	690 (590-810)	620 (510-720)	850 (220-1110)	1220 (1150-1260)
K^+	μg/l	590 (420-750)	430 (300-500)	380 (220-500)	430 (230-700)	350 (190-500)
Al_{tot}	μg/l	570 (340-680)	650 (550-730)	360 (280-500)	730 (550-970)	120 (90-150)

*) Chlorophyll *a*

Table 3. Selected nutrient parameters, phosphatase ativity, and chlorophyll *a* (all data for surface layer) of three Šumava lakes analyzed in 1992–1993.

Lake		Černé				Prášilské				Plešné			
Date		19-May 1992	19-Oct 1992	2-Aug 1993	25-Oct 1993	19-May 1992	20-Oct 1992	2-Aug 1993	25-Oct 1993	19-May 1992	20-Oct 1992	2-Aug 1993	26-Oct 1993
NH_4-N	µg/l	45	32	28	8	50	98	13	83	60	42	16	27
NO_3-N	µg/l	1180	970	970	935	1020	560	590	595	720	450	390	375
TN	µg/l	1360	1130	1230	1090	1330	870	990	890	1050	750	870	750
SRP*)	µg/l	1.0	2.9	<1	1.3	<1	<1	<1	<1	<1	<1	<1	
TP	µg/l	3.4	3.8	3.8	4.8	7.2	4.0	4.6	4.5	13.0	6.0	8.9	10.7
PP	µg/l			2.2	2.2	3.9	0.6	2.4	1.0	10.6	2.4	7.2	8.1
DOC	mg/l	1.8	1.1		1.1	3.7	3.4		4.0	3.7	1.7		2.4
POC	mg/l	0.44	0.56	0.60	0.36	0.71	0.89	1.3	0.36	0.24	1.9	1.9	1.8
POC:PP	molar			710	420	470	3800	1400	930	582	100	690	590
TN:TP	molar	880	660	720	500	410	480	480	440	180	280	220	160
PA*)	µmol/l h			0.85	0.84	0.67	0.79	1.3	0.68	0.59	4.8	13.4	8.0
Chl *a**)	µg/l	1.6		2.4	2.6	4.4		4.6	1.3	5.7		7.2	12.1
Z_s*)	m	11.0	8.0	7.6	7.8	3.9	7.0	2.6	5.8	2.9	2.6	1.7	1.8

*) SRP = soluble reactive phosphorus, PA = phosphatase activity, Chl *a* = chlorophyll *a*, Z_s = transparency.

4.99 were typical for other lakes (Table 2). Čertovo Lake was the most acidified one. Sulphate and nitrate were dominant anions. Increased concentration of Al_{tot}, which is typical for acidified fresh waters, was found in all lakes. A large portion of Al_{tot} was in the form of labile aluminium (FOTT & al. 1994). Concentration of nitrate suddenly dropped by ~ 30% in the early 1991s compared to the 1980s, while concentration of sulphate has continuously decreased since the 1980s (26%, see VESELÝ 1997 for more details). Consequently, pH slightly increased during the last decade. In 1993 and 1994, pH of the surface water in the lakes was by 0.15 to 0.3 unit higher than in the early 1980s, when historically the lowest pH values were observed (FOTT & al., pers. commun.).

The most prominent nitrogen form in lake water was NO_3-N. While hardly any difference was observed between spring and autumn NO_3-N concentrations in Černé Lake, it was by more than twice and nearly twice higher in May than from July to October in Plešné L. and Prášilské Lake, respectively (Table 3). The share of TON and NH_4-N in the TN ranged from 14% (Černé L.) to 42% (Plešné L.) and from <1% (Laka L.) to 7% (Plešné L.), respectively. The contribution of NO_2-N was less than 0.4%. Low concentrations of TON, COD and DOC were in Černé L. and Čertovo Lake, the high ones in Prášilské L., Plešné L. and Laka Lake (Table 2). In the latter lakes it resulted from an elevated input of allochthonous organic matter. Concentrations of COD in both seasonal and perennial inputs of surface water were < 5 mg l^{-1} for Černé Lake, while > 8 mgl^{-1} for Prášilské and Plešné Lakes. Higher TP concentrations found in Plešné Lake resulted from the elevated TP levels in the surface water inflows (usually between 10 to 30 µg l^{-1} in 1994). In contrast, substantially lower TP concentrations were found in the inflows into Černé L. and Prášilské Lake in 1992 and 1993; only from 1 to 4 µg l^{-1} and from 1.5 to 6 µg l^{-1}, respectively. A significant seasonality of NO_3-N concentrations, observed in the epilimnion of Plešné Lake, was related to phytoplankton development (due to higher TP input) and, consequently, more intense utilization of inorganic nitrogen.

All lakes (except for shalow Laka Lake) had typical summer thermal stratification. Differencies in stratification patterns of dissolved oxygen were observed among the lakes. While orthograde oxygen profiles remained in Černé L. and Čertovo Lake throughout the whole vegetative period, clinograde profiles with oxygen depletions (< 0.2 mg l^{-1}) above the bottom where typical for Prášilské L. and Plešné Lake during summer and winter periods. Hypolimnetic anoxia were accompanied by increased concentrations of NH_4-N, alkalinity, pH, and hydrogene sulphide, and by the decrease in NO_3-N and SO_4^{2-} concentrations. The extensive study of Plešné Lake in 1994 (KOPÁČEK & HEJZLAR, unpubl. data) showed strong vertical zonation of all parametres studied. Dissolved oxygen was exhausted in the bottom layer within one month after the beginning of thermal stratification. Then nitrate and later also sulphate started to be reduced, and, before autumnal circulation, less than 5% and < 60% of their spring overturn concentrations, respectively, remained in the water layer above the bottom. There also increased the concentrations of NH_4-N from 0.02 to > 1 mg l^{-1}, hydrogen sulphide from 0 to > 1 mg l^{-1}, iron from 0.25 to > 1 mg l^{-1}, and alkalinity from 3 to > 100 µeq l^{-1}. No orthophosphate was released from bottom sediments; elevated TP concentrations above the bottom resulted from resuspended particles from the upper layer of the sediment.

Considering the concentrations of nutrients, organic matter, and oxygen conditions, Černé L. and Čertovo Lake could be classified as oligotrophic while Prášilské L. and Plešné Lake as mesotrophic during the study.

Phytoplankton and primary production

Generally, by two to three orders of magnitude higher TN than TP concentrations (Table 2), and extremely high particulate organic carbon to particulate phosphorus (POC: PP) ratios of lake water (Table 3) indicated phosphorus limited phytoplankton growth in all lakes. This was also supported by the enhanced activities of phosphatase which were recorded during vegetation periods in all three lakes studied (Table 3). Despite the highest TP concentrations, the highest phosphatase activities (as high as 22 μmol l^{-1} h^{-1}) were observed in Plešné Lake probably due to substantially higher levels of phytoplankton biomass (1994's maxima of chlorophyll *a* reached up to 24 μg l^{-1} h^{-1}) compared to the other lakes. High phosphatase activities (but < 1 μmol l^{-1} h^{-1}) during phytoplankton maxima were also observed by MÜNSTER & al. (1992) in a polyhumic, phosphorus limited lake Mekkojärvi.

However, the extreme phosphatase activities recorded in our study cannot be solely explained by the low TP level and/or elevated phytoplankton biomass in lake water. They were most probably a phytoplankton response to blockade of organic phosphorus by the enhanced aluminium concentrations as suggested by JANSSON & al. (1986) and JOSEPH & al. (1995). The authors reported that dissolved inorganic aluminium forms may severely disturb the enzymatic regeneration of phosphate at low pH by blocking organic phosphorus compounds from enzymatic attack. Phytoplankton balanced this situation accordingly by producing more enzymes. Moreover, a similar effect of vanadium on algal phosphorus metabolism was described by NALEWAJKO & al. (1995). Vanadium may enter the lakes via atmospheric deposition, as it is commonly emitted from fossil fuel combustion processes, and may worsen the phosphorus acquisition, as well. VESELÝ & al. (1993) estimated an anthropogenic flux as 2.7 mg m^{-2} yr^{-1} of V into the sediment of Čertovo Lake.

Conspicuous vertical stratification of chlorophyll *a* was typical for both groups of lakes (not shown). Higher phytoplankton biomass was typical for the mesotrophic lakes (Plešné L., Prášilské L.), while concentrations of chlorophyll *a* did not exceed 3.7 μg l^{-1} in the surface water of Černé Lake (Table 2). However, local surface blooms of phytoflagellates have been observed in the oligotrophic lakes during a calm summer day (FOTT & al. 1994). As typical for acidified lakes, the phytoplankton of the Šumava lakes were also poor in number of species, being dominated by phytoflagellates (FOTT & al. 1994, LUKAVSKÝ in prep.). However, no picocyanobacteria typical for other acidified lakes (e.g. BELL & TRANVIK 1993, BELL & al. 1993) have been observed in the Šumava lakes.

Primary production as low as 0.98 to 1.17 mg C m^{-2} d^{-1} and 3.05 mg C m^{-2} d^{-1} was determined in Černé L. and Prášilské Lake, respectively, during the vegetation period in 1991. The results are mostly higher than those observed by JANSSON & al. (1986) in acidified lakes, but one order magnitude lower than those in polyhumic Mekkojärvi (SALONEN & JOKINEN 1988). More intense primary production could be excepted in Plešné Lake where the chlorophyll *a* concentrations are higher. Our results suggest that phosphorus metabolism of the Šumava lakes may be also disturbed due to acidification which may impair the primary production (cf. JANSSON & al. 1986).

Zooplankton

While crustacean zooplankton were very rich and abundant in the Šumava lakes 120 years ago (for review, see VESELÝ 1994), in the 1930s, their abundances and species composition were reduced due to unclear reasons, and later crustacea almost disapeared due to acidification (for Černé Lake, see Table 4; FOTT & al. 1994, PRAŽÁKOVÁ & FOTT 1994). During this study, rotifers were scarce in all lakes and planktonic crustacea were missing in three lakes (Černé L., Čertovo L., Plešné Lake), whereas herbivorous filtrators (*Daphnia longispina*) and copepods (*Cyclops abyssorum*) were regularly recorded in Prášilské Lake. FOTT & al. (1994) explained this difference with distinct aluminium concentrations among the Šumava lakes. While pHs were the same, concentrations of Al_{tot} were about twice lower in Prášilské Lake compared to the others (Table 2). The percentage of the most toxic, labile forms of aluminium is also a bit lower in Prášilské Lake (FOTT & al. 1994).

Table 4. – Planktonic crustacea in Černé Lake, from 1871 to our times (according to Fott et al. 1994)

Period	1871 ~	1892 to 1896	1935 to 1937	1947 ~	1960 ~	1979 to 1989
Holopedium gibberum	+	+		+		
Daphnia longispina	+	+	*			
Ceriodaphnia guadrangula		+	+	+	+	**
Bosmina longispina	+	+				
Acanthodiaptomus denticornis	+					
Cyclops abyssorum	+	+	+	+	+	

* 2 specimens found in 1935, ** 1 specimens found in 1979, ~ one sample

Table 5. – Microbiological parameters (mean, range in parentheses) of three Šumava lakes during 1990–1994 (different number of samples, data from both surface and bottom layers are included)

Parameter	Černé L.	Prášilské L.	Plešné L.
Bacteria (10^6 cell ml^{-1})	0.40 (0.06–0.80)	1.09 (0.15–2.68)	1.72 (0.51–6.90)
Bacterial biomass (10^6 µm^3 l^{-1})	35 (3–129)	66 (5–149)	102*)
α-glucosidase**) (amol cell^{-1} h^{-1})	4.0 (0.7–8.9)	2.0 (0.2–10.2)	1.7 (0.3–5.1)
β-glucosidase**) (amol cell^{-1} h^{-1})	12.8 (3.7–19.0)	9.9 (1.3–35.3)	4.7 (0.9–12.4)
β-hexosaminidase**) (amol cell^{-1} h^{-1})	21.0 (1.9–64.6)	3.1 (0.8–10.0)	3.7 (0.7–14.3)
HNF (cell ml^{-1})	307 (67–690)	539 (40–2369)	389 (19–1780)
Mean cell volum of HNF (µm^3)	68 (24–210)	86 (6–454)	52*)
Protistan grazing (10^3 bact. ml^{-1} h^{-1})	1.1*)	12.3*)	21.8 (6.2–48.6)

*) only one measurement, **) specific ectoenzyme activities per bacterial cell

Bacterioplankton

Bacterial abundances were, on average, 2 to 4 times higher in the mesotrophic than in the oligotrophic lakes (Table 5). Average bacterial biomass in Prášilské L. was about twice as high as in Černé Lake. The minimum and maximum values were, however, almost the same (Table 5) because larger bacteria usually dominated in Černé Lake (see mean cell volumes in Table 6). Besides filamentous bacterial cells in zooplanktonless lakes, bacterial biomass was also enhanced by large and abundant bacteria in anoxic or anaerobic strata of the hypolimnion of both mesotrophic lakes. The bacterial biomass was comparable with another acidified lake Gardsjön (Jansson & al. 1986). Bacterial counts in all mesotrophic lakes were almost the same as in an oligotrophic lake Njupfatet (pH ~ 5.7) from central Sweden (Bell & al. 1993) and also in the polyhumic Mekkojärvi (Salonen & Jokinen 1988), while bacte-

Table 6. – Comparison of some microbiologial parameters (bacteria: number = BN in 10^6 cell ml^{-1}, mean cell volume = BCV in μm^3, biomass = BB in 10^6 μm^{3}l^{-1}: HNF: number = HNFN in cell ml^{-1}, mean cell volume = HNFCV in μm^3, biomass = HNFB in 10^6 μm^3 l^{-1}) in two Šumava lakes during one vegetation period (1991). Items at distinct, i.e. epi - (surface) and hypolimnetic (above bottom or at 26 m depth) temperatures are compared in absence (A) or presence (P) of crustacean zooplankton (ZOO).

	BN	BCV	BB	HNFN	HNFCV	HNFB	ZOO
Černé L. – May							
surface	0.35	0.085	30	265	36	10	A
26 m	0.24	0.054	13	192	210	40	A
Černé L. – October							
surface	0.49	0.265	129	216	25	5	A
26 m	0.43	0.168	73	67	32	2	A
Prášilské L. – May							
surface	0.55	0.096	53	251	46	11	A
bottom	0.75	0.104	77	150	126	19	A
Prášilské L. – Oct.							
surface	0.96	0.063	60	468	16	7	(P)
bottom	2.33	0.049	115	72	6	0.5	P

rial abundance in both oligotrophic lakes was less than half those in Njupfatet. However, as bacteria from Černé Lake were usually several times larger than those in Mekkojärvi (Salonen & Jokinen 1988), bacterial biomass met the same range in the both lake compared.

In addition, we estimated bacterial production in two lakes during the vegetation period of 1991. The thymidine incorporation was twice higher in the mesotrophic Prášilské Lake (mean: 8.04, range: 1.79–14.42 pmol l^{-1} h^{-1}) compared to the oligotrophic Černé Lake (mean: 4.04, 0.63–8.42 pmol l^{-1} h^{-1}). The maximum values were even one order of magnitude higher than those from lake Njupfatet with comparable or higher bacterial abundances (Bell & al. 1993). This comparison suggested that pelagic bacteria were likely more productive in the Šumava lakes. One could conclude that bacterial production was roughly proportional to bacterial biomass in the lakes studied. On the other hand, bacterioplankton of Černé Lake was more active on a per-cell basis compared to both mesotrophic lakes: e.g., per-cell ectoenzymatic activities were always the highest in Černé Lake (Table 5).

Ectoenzyme activities

To estimate microbial decomposition of DOC and POC, we measured a degradation of common polysaccharides by bacterial ectoenzymes. Activities of α-glucosidase, β-glucosidase, and β-hexosaminidase are considered as the final steps in hydrolysis of starch, cellulose, and both chitin and peptidoglycan, respectively. Total activities of these enzymes in lake water are given in Table 7. Dissolved activities (in 0.2 μm filtrate) accounted for 0–75% (α-glucosidase), 0–92% (β-glucosidase), and 4–99% (β-hexosaminidase). Results on α- and β-glucosidases met those observed in eutrophic and polyhumic waters (Münster 1991, Vrba 1992). However, maximum β-hexosaminidase activities in Černé L. and Plešné Lake (Table 7) were surprisingly higher than those from a eutrophic reservoir (Vrba 1992) while neither crustacean zooplankton nor diatoms (i.e. potencial substrate sources) were present in the lakes. One could speculate about occurrence and moulting of water insect larvae or some allochthonous amino sugar supply from the basins. Another possible source of a high-affinity β-hexosaminidase activity may be protistan grazing (Vrba & al. 1996). We actually observed protistan bacterivory in the lakes, however, the high-affinity β-hexosaminidase activity probably did not exceed 1 nmol l^{-1} h^{-1} (Fig.2, see also below).

Table 7. – Total extracellular activities (mean, range in parentheses) of α-glucosidase, β-glucosidase, α:β ratio, and β-hexosaminidase (in nmol l^{-1} h^{-1}) in three Šumava lakes during 1991–1994.

Lake	α–glucosidase	β-glucosidase	α:β	β-hexosaminidase
Černé	2.0 (0.3–4.4)	6.0 (2.2–9.4)	0.33	9.8 (0.8–29.2)
Prášilské	1.9 (0.3–5.0)	8.4 (1.4–22.2)	0.46	2.8 (0.5–4.9)
Plešné	3.2 (1.0–7.8)	8.6 (4.6–18.7)	0.36	8.3 (1.8–33.8)

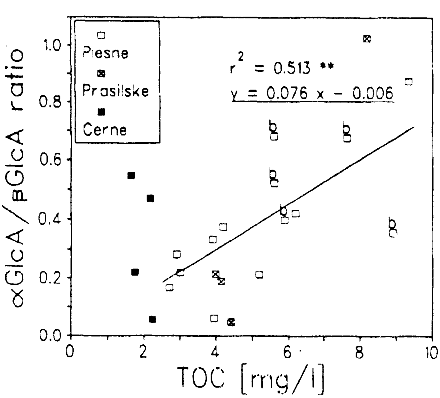

Fig. 1. Relationship between total organic carbon (TOC = DOC + POC) and the ratio of α-glucosidase (αGLcA) to β–glucosidase (βGlcA) activity. The linear regression was calculated only for Plešné data (b=sample taken above the bottom).

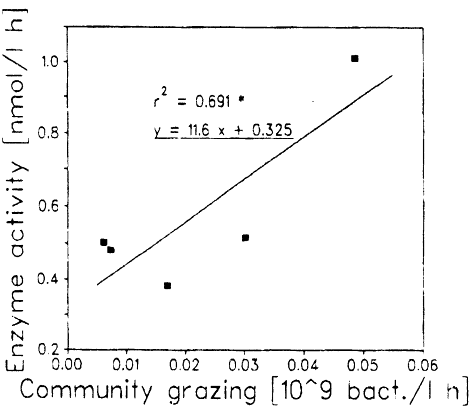

Fig. 2. Relationship between protistan bacterivory (community grazing estimated via fluorescently labelled prey) and protistan digestive activity (maximum velocity of the high-affinity β-hexosaminidase distinguished kinetically, see Vrba et al. 1996 for more details).

Ratios of α- to β-glucosidase activity were mostly very low (Table 7, Fig.1) indicating likely a degradation of recalcitrant plant material (DOC plus POC) from the basins (Münster 1991, Vrba 1992) of all lakes. A similar low ratio of α:β was found by Münster (1991) for all glycosidic ectoenzymes in polyhumic Mekkojärvi. The average ratio of α-:β-glucosidase in Mekkojärvi (0.39) just fitted the mean values from the Šumava lakes (Table 7). We found a significant correlation ($r^2 = 0.513$) of the α-:β-glucosidase ratio and the sum of DOC plus POC (Fig.1), but neither for sole DOC nor POC concentrations in Plešné Lake. Surprisingly, high POC of unknown nature in the samples taken above the bottom induced somewhat enhanced α-glucosidase activities.

Protozoa

On average, HNF were less abundant in the oligotrophic than in the mesotrophic lakes (Table 5) and ciliates, although in very low numbers, were observed in all lakes (Macek, unpubl. data). Mean cell volumes of HNF showed a characteristic, inverse relation to water temperature, i.e., increasing cell volumes towards a bottom of each lake. Both low temperature and hypoxia were found to favour the occurrence of larger protozoans in the hypolimnion of a eutrophic lake (Sime-Ngando & Hartmann 1991). Cladoceran predation broke this pattern whenever a population of *D. longispina* appeared in any layer of Prášilské Lake which usually resulted in minimum abundances of, moreover, very tiny HNF. In anoxic layers of Prášilské Lake, the daphnids could not survive and HNF remained large up to the end of summer stratification when anoxia-tolerant copepods (*C. abyssorum*) appeared in hypolimnion

(STRAŠKRABOVÁ & ŠIMEK 1993). The HNF population then was grazed out to a very low abundance and biomass (Table 6). On the other hand, maximum abundances of HNF (Table 5) usually coincided with some peaks of chlorophyll *a* in both mesotrophic lakes. The highest mean HNF abundance was found in Prášilské Lake, however, it was almost three times lower compared to that of polyhumic Mekkojärvi (SALONEN & JOKINEN 1988).

Microbial loop interactions

Protistan community grazing on bacterioplankton was more than one order of magnitude higher in both mesotrophic lakes compared to Černé Lake (Table 5). Our results fitted the general relationship between total protistan bacterivory and activity of the high-affinity, β-hexosaminidase-like, protistan digestive enzyme (Fig.2, VRBA & al.1996). In all lakes, however, pigmented flagellates (e.g., dinophytes or colonial chrysophytes, *Dinobryon* spp.) were responsible for as much as 40% of the community grazing. Similar results and even higher share of mixotrophic flagellates on total bacterivory were frequently reported from oligotrophic and/or acidified lakes of Northern Hemisphere (BELL & TRANVIK 1993 and references therein) and also from polyhumic Mekkojärvi (SALONEN & JOKINEN 1988). Proportion of filaments, i.e., forms considered as grazing-resistant to protozoa (JÜRGENS & GÜDE 1994), was usually higher within bacterioplankton of the lakes without cladocerans. In contrast, no bacterial filaments were recorded in Prášilské Lake when HNF (and probably also filaments) were grazed out by zooplankton (compare the mean cell volumes of bacteria in October 1991, Table 6).

Our results support a general tendency that acidification itself does not induce the major physiological stress of bacteria (BELL & TRANVIK 1993). Relatively high bacterial production compared to limited primary production, profiting also from supply of allochthonous recalcitrant DOC, is cropped by heterotrophic and mixotrophic protists. On the other hand, HNF populations, differing strongly in species composition between the epi- and hypolimion, are apparently affected by temperature and also by availability of bacterial or other prey (bottom-up control).

As higher trophic levels are mostly absent due to acidification, a unique type of aquatic ecosystem, with a dominance of microbial processes in pelagic food webs, creating so-called microbial loop (Fig. 3), has developed in the Šumava lakes. As far as we know, such type of acidified lakes has not yet been described in the world. Except for Prášilské Lake, the only potential herbivores could be some larger protozoa and scarce rotifers. Thus, most of primary production as well as allochthonous organic matter are utilized by heterotrophic bacteria and colourless (HNF) or mixotrophic flagellates. The latter play also an important role in nutrient recycling towards primary producers. All conclusions fit appropriately the TRANVIK's (1992) concept of the microbial loop in acidic and humic lakes.

Our preliminary results suggest that mixotrophy (phagotrophy) of pigmented flagellates covers a significant portion of nutrient and/or energy requirements of phytoplankton in the Šumava lakes. As bacterial cells are relatively rich in phosphorus compared to phytoplankton, the phagotrophy may be an alternative pathway to cover algal demand for P (SALONEN & JOKINEN 1988, BELL & TRANVIK 1993) under an acidification stress when extracellular phosphatases are blockaded (see above). The latter might be one of the factors responsible for influencing species composition of phytoplankton in acidifed lakes.

The top-down effect of daphnids (and also of cyclopids) on HNF populations was evident in Prášilské lake. The herbivorous filtrators could have compensated for a low primary production by feeding on HNF. Thus, even in the presence of crustacean zooplankton, the microbial loop forms an important link through which allochthonous DOC could be imported to higher trophic levels of the pelagic food webs as suggested by TRAVNIK (1992).

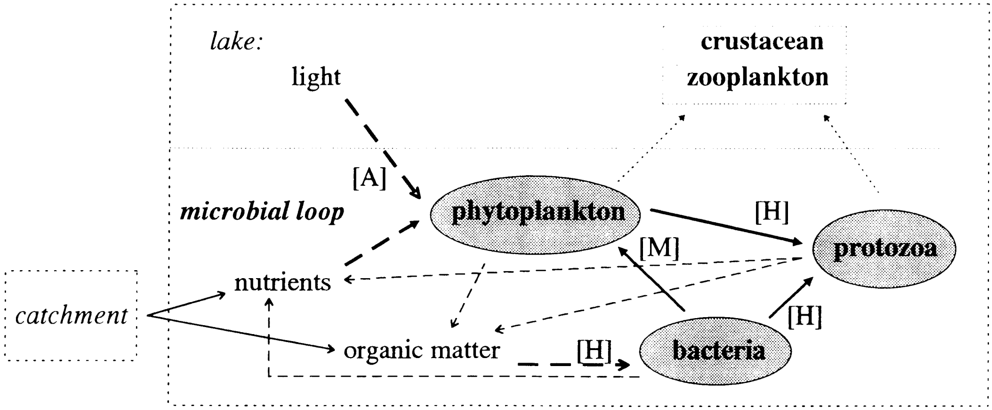

Fig. 3. – A scheme of pelagic food web interactions, carbon flow and nutrients: [A] – autotrophy, [H] – heterotrophy, [M] – mixotrophy. Dominance of the microbial loop (lower part) is typical for all Šumava lakes. Higher tropic levels (top) are only temporaly present in Prášilské lake.

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