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HORIZONTAL DISTRIBUTION OF CILIATED PROTOZOA BETWEEN THE SPHAGNUM MAT AND OPEN WATER ZONE IN A SHALLOW PEAT-BOG POOLS[1]

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Summary. The aim of this study was to examine the community structure and horizontal micro-distribution (under the peat mat, transitional zone peat mat/open water and open water zone) of ciliates in shallow peat-bog pools in eastern Poland, as well as to assess the effect of physical and chemical factors on the distribution of planktonic ciliates. The highest species richness occurred in the transitional zone. Decidedly lower numbers of taxa were observed in the open water zone. The density and biomass of protozoa significantly differed between the studied zones, with the lowest numbers in the open water zone and the highest between peat mat and open water. Analyses at the level of species and genera revealed distinct distribution patterns of some taxa. *Strombidium viride* and *Vorticella companula* as medium-sized showed highest abundance in open water. Small and medium-sized *Cinetochilum margaritaceum* and *Colpoda* sp. were highly abundant in the transitional and in the peat mat zones. In the peat mat, as well as in contact zone, the number of ciliates had the strongest correlation with water temperature, conductivity, concentrations of total organic carbon. In turn, in the open water zone, there was a significant rise in the strong correlation between the number of ciliates and the total phosphorus concentration.

Key words: peatbog, pools, ecotone, Sphagnum, ciliates

INTRODUCTION

Small peat-bog pools with low pH, high concentrations of humic matter have low levels of inorganic nutrients. The typical brown water colour reduces light penetration, thus primary productivity is usually low. Low pH is a biologi-

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cal effect of peat-moss occurring around a reservoir or even expanding over its water table from of a mat with water pH about 3.0–5.0 [Tranvik 1988]. Pool micro-habitats are regarded as hotspots for biodiversity in peatbogs. Both generalist and specialist aquatic insects are found here, and the intrinsic variety of aquatic invertebrates supports vertebrate diversity via trophic links. It is generally assumed that the topographical and hydrological characteristics of peatbog pools influence not only faunal diversity, but also floral diversity [Fontaine *et al.* 2007]. The littoral zone in peat-bog pools comprises a mosaic of vertical and horizontal microhabitats, provided by Sphagnum mosses and open patches [Henrikson 1993]. These habitats generally reflect a very diverse composition with successive development being essential due to emergent and submerged vegetation that comprises a number of life forms. Macrophyte beds are favourable for zooplankton development, owing to the refuge they provide against fish predation. Many studies have reported that in the presence of planktivorous fish, zooplankton abundance is much higher within dense plant beds than in open water [Basu *et al*. 2000]. In small peat-bog reservoirs the animal communities, especially invertebrates, are sufficiently known [Błędzki and Ellison 2003]. By contrast, little or no attention is given to the abundance and biomass of protozoans and their relationships to micro-site differentiation in these specific ecosystems. These microorganisms are important consumers of bacteria, flagellates and algae; they also participate in the decomposition of organic matter and nutrient cycling. Ciliates are a group of protozoa that have been shown to be good indicators of hydrology and water chemistry in lakes and peatlands [Packroff 2000, Mieczan 2005]. However, there is still not enough information available on the species segregation in shallow peat-bog reservoirs. To date, no research has been carried out on planktonic ciliates in the transitional zones between the peat mat and water, and in relation to the some physical and chemical parameters of peat-bog pools. The aim of the present study, therefore, was to establish the following: whether differences exist between planktonic ciliate communities in different zones (under the peat mat, transitional zone peat mat/open water and open water zone), and to assess the effect of physical and chemical factors on the distribution of planktonic ciliates in three shallow peat-bog pools.

STUDY AREA, MATERIAL AND METHODS

Studies have been carried out in three small peat-bog pools (area < 0.5 ha, mean depth 0.9 m). The reservoirs are located in Jelino peatbog (Łęczna--Włodawa Lakeland, Eastern Poland) and have been created as a result of peat extraction. Most of the reservoirs adjoin a peatbog formed by Sphagnum and covered by other plants characteristic of peatbogs: *Carex acutiformis* Ehrhart. *Carex gracilis* Curt., *Equisetum limosum* (L.), *Sphagnum* sp. and *Polytrichum* sp. The study was carried out from April to November 2008, in the shallowest part of the peat-pond pools, at the border of the peat mat overgrowing the water

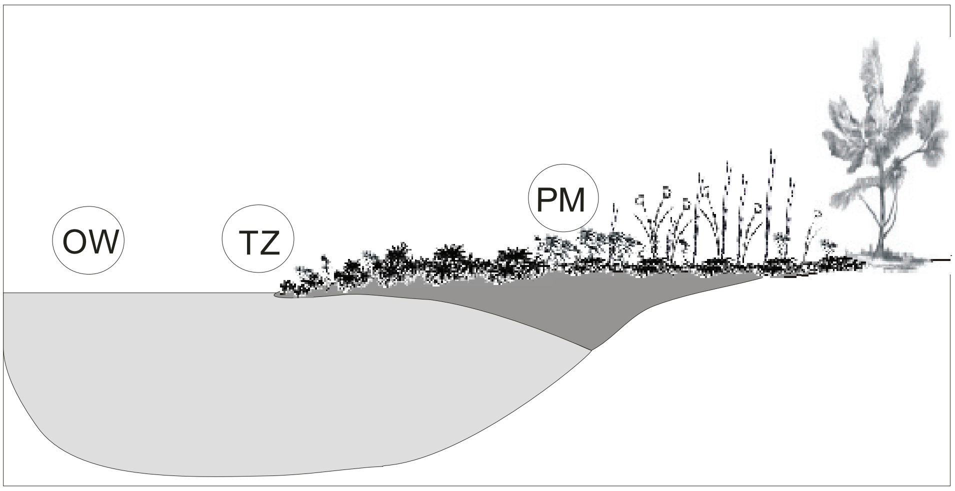

Fig. 1. Location of the sampling points (PM – peat mat, TZ – transitional zone between the peat mat and open water zone, OW – open water zone)

surface. The samples (each in triplicate) were collected from three stations: PM – under the peat mat, TZ – the transitional zone between the peat mat and open water zone, OW – the open water zone (Fig. 1). At each type of zone and each sampling date water was sampled using a plexiglass corer (length 1.0 m, Ø 50 mm). The plexiglass corer was closed at each end with a cork and then water samples were collected using a glass pipette. Protozoa samples (whole sample = 500 ml) were sedimented for 24 h in cylinder, stoppered with parafilm, then the upper volume of 400 ml was gently removed. In order to determine the density and biomass, two samples were preserved with Logol solution (0.2% final concentration). Observation of live samples was used for the taxonomic and trophic identification. Morphological identifications of the ciliates were mainly based on works by Foissner and Berger [1996] and Foissner *et al.* [1999].

The water samples for chemical analysis were taken simultaneously with the plankton samples. The following physical and chemical factors were examined: temperature, pH, conductivity, total organic carbon (TOC), phosphates and total phosphorus. Temperature, conductivity and pH were recorded *in situ*. TOC was determined using the PASTEL UV; the remaining factors were analysed in the laboratory [Hermanowicz *et al.* 1976].

Diversity analysis (Shannon-Wiener diversity index) was performed using the Multivariate Statistical Package (MVSP) [Kovach Computering Services, 2002].

The frequency of occurrence of a particular species was calculated as a percentage of collected samples in which the species occurred. All species were classified into 4 groups as follows: very constant species (occurring in 61–100% of samples); constant species (occurring in 41–61% of samples); accidental species (occurring in 21–40% of samples); accessory species (occurring in less than 20% of samples).

The similarity of planktonic communities between the zones was calculated by the Jaccard method:

$$S_{xy} = \frac{c}{a + b - c} \cdot 100\%$$

where:
S_{xy} – faunistic similarity between data sets x and y,
c – number of taxa common for sets x and y,
a – number of taxa in set x,
b – number of taxa in set y.

All data collected were statistically analysed by means of GLM and CORR procedures of the SAS Programme [SAS Institute Inc. 2001]. One-way ANOVAs with *post-hoc* Bonferroni tests were run on abundance data to assess separately the protozoan variability caused by the reservoirs and the zones (n = 52). Correlation between physical and chemical parameters and ciliate density were analysed by calculating Pearson's correlation.

RESULTS

Statistically significant differences between individual zones in the peat-bog pools studied were indicated in water temperature, pH, conductivity and the concentration of total organic carbon (p = 0.0111–0.0201). The next significantly high concentration was in nutrient (PO_4 and P_{tot}) found in the open water zone (p = 0.0301). In the three reservoirs examined, the water temperature reached the highest value in the peat mat (18.8–19.6°C), and decreased in the direction of the open water zone (14.0–15.3°C). Conductivity decreased in the direction of the open water zone and ranged from 44.8 to 27 µS cm^{-1}. Likewise, the TOC content significantly rose in the transitional zone between the peat mat and open water zone (24.7–25.3 mg C dm^{-3}) and decreased in the direction of the open water zone (10.4–10.8 mg C dm^{-3}). Concentrations of P_{tot} and P-PO_4 were mostly low in the three reservoirs, and showed an increasing tendency along the transect with the maximum in the open water zone (Tab. 1).

A total of twenty four species were found in the studied peat-bog pools. The number of ciliate taxa was similar in the three studied pools, but revealed a statistically significant difference between the investigated zones (p = 0.0021). The highest richness was found in the transitional zone between the peat mat and open water (18) and the lowest in the open water zone (9). In the open water contact zone *Strombidium viride* was a very constant species; in the 2 remaining zones *Cinetochilum margaritaceum* and/or *Colpoda* sp (Tab. 2). The diversity analysis revealed a mean Shannon-Wiener diversity index of 1.2. The highest diversity was measured in the transitional zone (H = 1.9) and the lowest diversity was observed in open water zone (H = 0.83). The Jaccard similarity index reached a mean value of 67–80%. The mean numbers of planktonic ciliates changed

Table 1. Physical and chemical characteristics of water in the investigated peat-bog pools and zones

Peat-bog pools	Zone	Temp., ºC	pH	Contuctiv., $\mu S \cdot cm^{-1}$	PO_4, $mg \cdot dm^{-3}$	Ptot, $mg \cdot dm^{-3}$	TOC, $mg \cdot dm^{-3}$
1	PM	18.8	2.91	41.5	0.180	0.200	26
	TZ	14.7	4.27	44.8	0.127	0.154	25.3
	OW	14.2	4.93	26.3	0.461	0.276	10.4
2	PM	19.6	2.91	41.3	0.191	0.211	23
	TZ	15	4.1	42.5	0.109	0.17	25
	OW	14	4.5	27	0.411	0.222	10.8
3	PM	19.3	3.2	40.2	0.163	0.196	20.7
	TZ	14.7	4	43.2	0.123	0.177	24.7
	OW	15.3	4.17	28.7	0.460	0.274	10.8

PM – peat mat, TZ – the transitional zone between the peat mat and open water zone, OW – the open water zone; average values April–November 2008

Table 2. The frequency of occurrence of a particular ciliates species in the investigated zones

Taxa/zone	PM	TZ	OW
Very constant	*Colpoda sp.*	*Colpoda sp.* *Cinetochilum margaritaceum*	*Strombidium viride*
Constant	*Cinetochilum margaritaceum*	*Strombidium viride* *Spathidium sensu lato* *Paramecium bursaria*	*Vorticella companula* *Paramecium bursaria* *Codonella cratera*
Accidental	*Paradileptus elephantinus* *Paramecium bursaria* *Spirostomum ambigum* *Prorodon sp.*	*Colpidium colpoda* *Paradileptus elephantinus* *Spirostomum ambigum* *Cinetochilum margaritaceum* *Litonotus sp.* *Prorodon sp.* *Chilodonella uncinata*	*Askenasia volvox*
Accessory	*Trochilia minuta* *Askenasia volvox* *Trachelius ovum* *Cyclidium citrulus* *Codonella cratera* *Kahlilembus attenuotus*	*Askenasia volvox* *Trachelius ovum* *Cyclidium citrulus* *Codonella cratera* *Cyrtohymena muscorum* *Kahlilembus attenuotus*	*Colpoda sp.* *Lacrymaria olor* *Aspidisca costata* *Coleps hirtus*

PM – peat mat, TZ – the transitional zone between the peat mat and open water zone, OW – the open water zone.

in the individual zones. In the three pools, there was a significantly higher abundance of ciliates prevalent in the transitional zone between peat mat and open water (from 21 to 27 ind. ml^{-1}) and decreased in the direction of the open water zone ($p = 0.012$). The lowest biomass of ciliates was noted in the open water zone – 11 µg ml^{-1}. In the other two zones, the biomass of ciliates fluctuated from 13–14 in the peat mat to 16–18 µg ml^{-1} in the peat mat/open water contact zone (Fig. 2). In the peat mat/open water contact zone,

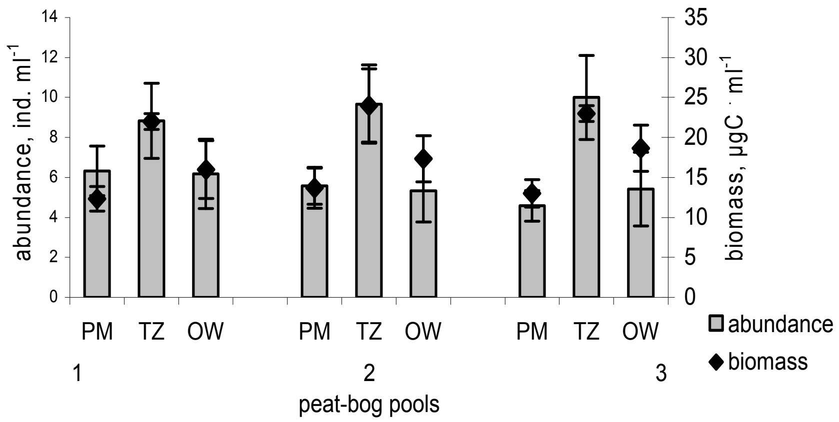

Fig. 2. Average density and biomass of planktonic ciliates in the investigated peat-bog pools and zones; PM – peat mat, TZ – transitional zone between the peat mat and open water zone, OW – open water zone; average values April–November 2008 ±SD

as well as in the peat mat, the number of ciliates had the strongest correlation with water temperature ($r = 0.73–0.75$, $p < 0.01$), conductivity ($r = 0.53–0.59$, $p < 0.01$) and concentrations of TOC ($r = 0.86–0.90$, $p < 0.01$). In turn, in the open water zone, there was a significant rise in the strong correlation between the number of ciliates and the total phosphorus concentration ($r = 0.33–0.37$, $p < 0.05$).

Among the four dominating species of the investigated peat-bog pools (*Strombidium viride*, *Cinetochilum margaritaceum*, *Vorticella companula* and *Colpoda* sp.) different patterns of spatial distribution were observed. Numbers of *Cinetochilum margaritaceum* and *Colpoda* sp. decreased from the central part of the peat bog towards the open water zone, while the distribution of *Strombidium viride* and *Vorticella companula* showed the opposite pattern (Fig. 3a–d).

Ciliates combined from all sampling dates and sites were dominated by medium-sized ciliates (50–200 µm) comprising of up to 50–90% of the total. Small species (15–50 µm) represented 10–45% and large ciliates (> 200 µm) 10–23% of the samples. Size classes of ciliates clearly differed between the individual zones. Peat mat zones were dominated by large forms; whereas, the remaining zones were dominated by small and medium ciliates. Ciliate feeding groups consisted of bacterivores, algae-diatom feeders, mixotrophic ones, predators, and omnivores. Bacterivore taxa clearly dominated in the transitional zone – peat mat/open water. In turn, the open water zone and peat mat were dominated by omnivorous and mixotrophic ciliates, at 36–45% of the total number (Fig. 4).

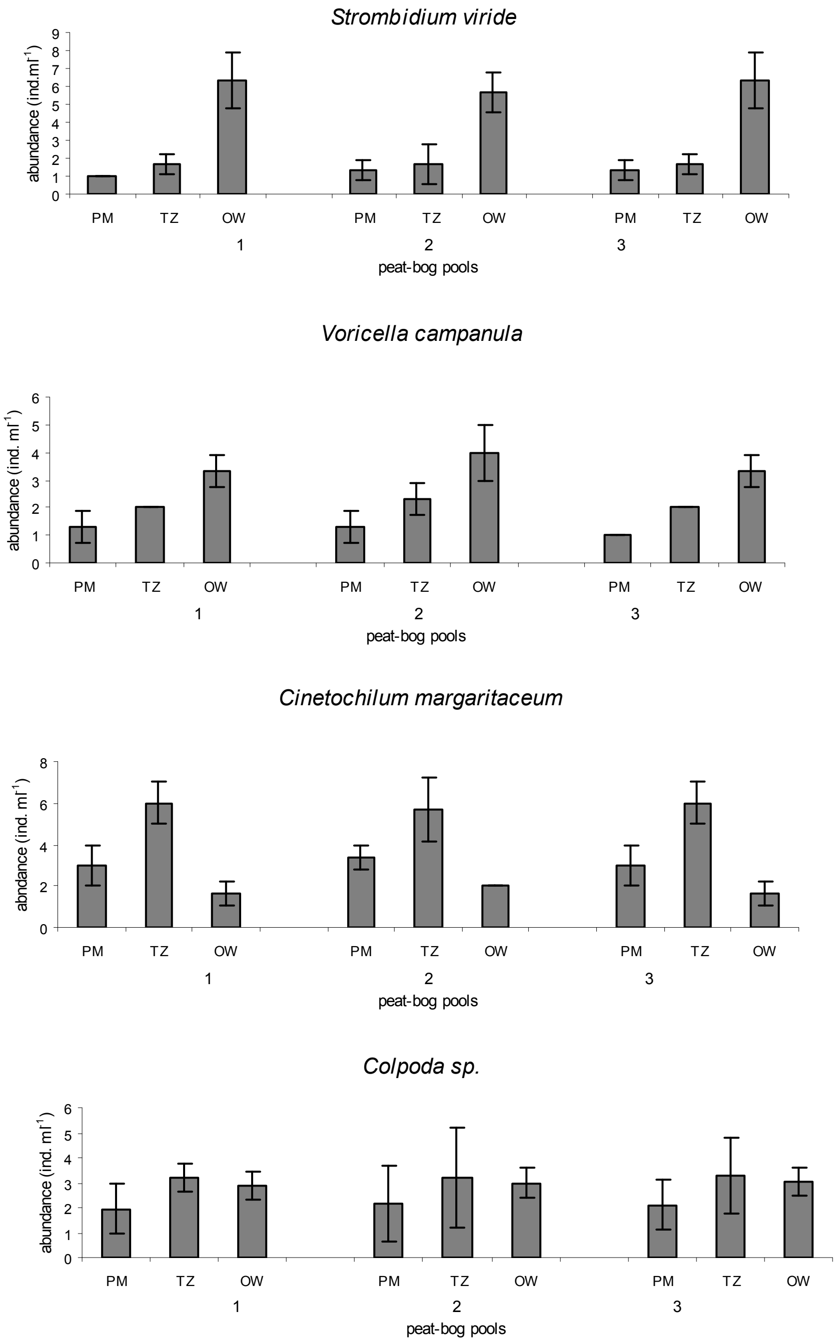

Fig. 3. Mean densities of the dominating taxa of ciliates in the investigated peat-bog pools and zones; PM – peat mat, TZ – transitional zone between the peat mat and open water zone, OW – open water zone; average values April–November 2008 ±SD

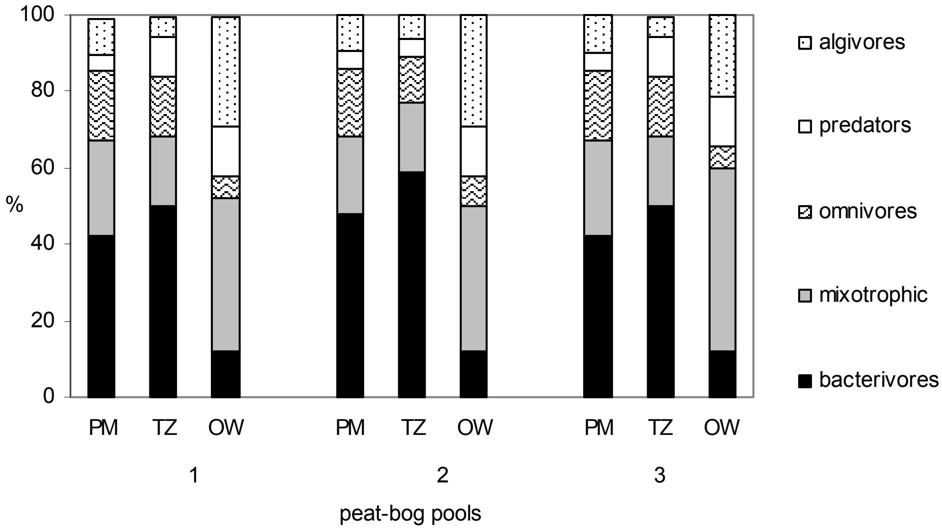

Fig. 4. Percentages of dominant feeding groups of ciliates in the investigated peat-bog pools and zones; PM – peat mat, TZ – transitional zone between the peat mat and open water zone, OW – open water zone

DISCUSSION

Ecotone zones (including the transitional zone between the peat mat and the open water zone) in peatbogs have not been investigated sufficiently so far. Therefore, they are increasingly coming into the focus of researchers [Beech and Landers 2002]. The function of these zones in shaping the species diversity of protozoa is particularly little known. In the investigated peatbog pools, significantly higher number of taxa and diversity of ciliates in the transitional zone between the peat mat and the open water zone was found. It is probably a result of „the edge effect" – the occurrence of the transitional zone, often characterised by growth in the species richness and abundance [Neiman and Decamps 1997]. Moreover, it seems that the abundance of ciliate species in the zones analysed could be, to some extent, shaped by the physical and chemical properties of water. A definite increase in conductivity, as well as higher content of total organic carbon, were probably among the causes for such species profusion. Results of other surveys also prove an increase in taxonomic richness with an increase of total organic carbon concentrations and conductivity [Mieczan 2007a, 2010]. Also the reaction of water may have a significant impact on the taxonomic composition of planktonic ciliates. Numerous surveys prove that individual taxa are characterised by different tolerance ranges for this parameter [Weisse and Stadler 2006]. This study reports a decrease in the species diversity along with a decrease in pH. Also Crisman and Brezonik [1980], as well as Mieczan [2007a], confirmed that an increase in water acidity is accompanied by a decrease in the species diversity of these microorganisms.

Available literature does not provide any comparative information concerning the taxonomic composition of planktonic ciliates in transitional zones of peat-bog pools. Much more attention is paid to ciliates from humic lakes [Beaver and Crisman 1989, Carrias *et al.* 1994, Packroff 2000, Graham *et al.* 2004, Tadonleke *et al.* 2005]. However, those surveys were restricted to the open water zone only. In the humic Lake Lisunie in the Masurian Lake District, the same number of taxa (17) [Kalinowska 2000] was found as in the open water zone in the peat-bog pools studied. It was also observed that in lakes with pH of approximately 5 or less, the coastal zone was richer in species than the open water zone [Kalinowska 2000]. Moreover, high values of the similarity index among individual zones probably resulted from the fact that most ciliates are organisms of wide ecological tolerance and significant easiness of spreading throughout the ecosystem [Finlay *et al.* 1999, Esteban *et al.* 2000].

The numbers and biomass of ciliates showed significant zonal variation. The highest density and biomass of these microorganisms was observed in the transitional zone between the peat mat and the open water zone, and the lowest in the open water zone. A study carried out by Beech and Landers [2002] shows that changing environmental conditions may affect the growth of the abundance of ciliates in the transitional zone. As demonstrated in several earlier studies, the abundance of these microorganisms is positively correlated with TOC, conductivity, and content of organic matter in the water [Beaver and Crisman 1989, Sarvala *et al*. 1999, Mieczan 2005, 2007a, c]. The study also indicates a clear positive correlation between the abundance of planktonic ciliates and the contents of total phosphorus in the peat-bog pools studied. Beaver and Crisman [1981] demonstrated that abiotic parameters may have a stronger impact on the abundance of ciliates than the trophic parameters of the environment (abundance of bacteria and „fine" phytoplankton). It also seems that planktonic ciliates may use the transitional zone between the peat mat and the open water zone as a potential refuge from the predation pressure of zooplankton (rotifers and planktonic crustaceans).

In all the peat-bog pools studied, both in the peat mat zone and in deeper parts of the waters, the most numerous species was *Strombidium viride* (Oligotrichida). Large numbers of Oligotrichida were also observed in a few peat-bog pools of the Polesie Lubelskie Region, and in humic lakes [Pace 1982, Carrias *et al.* 1994, Kalinowska 2000, Graham *et al*. 2004, Mieczan 2007a]. Research on ciliates in the Łęczna-Włodawa Lakeland showed that in peat-bog pools with lower pH and higher TOC concentration mainly Oligotrichida (*Strombidium viride*) occurred. The abundance of Scuticociliatida (*Cinetochilum margartaceum*) increased along with an increase in pH [Mieczan 2007a]. In the transitional zone, in addition to Oligotrichida, ciliates from Colpodea, mostly *Colpoda* sp., predominated. This taxon is strictly related to moss, which explains its abundance in the transitional zone between the peat mat and the open water zone. Microhabitats with the predominance of *Sphagnum palustre* were also dominated by Colpodea [Mieczan 2007b].

Regardless of the biotic zone, the investigated peat-bog pools were dominated by omnivorous and bactivorous taxa. A similar situation was observed in a few peat-bog pools in the Łęczna-Włodawa Lakeland, where bactivorous taxa and omnivorous taxa represented 35–60% and 20 to 46% of the total numbers, respectively [Mieczan 2007a]. Both in the transitional zone between the peat mat and the open water zone, and in the peat mat itself, a large number of bactivorous taxa was found. This is probably related to the conditions occurring in this zone. Decaying remains of plants favour the massive occurrence of bacteria, which triggers a violent growth of bactivorous ciliates. High concentration of humic matter in the peat-bog pools may also affect the amount of bacterial nutriment [Carlsson *et al.* 1995]. The least represented of all trophic groups were algivore ciliates. This could result from higher concentration of humic matter in the peatbog pools. These compounds „compete" with phytoplankton, which results in a reduction of light penetration and, as a consequence, a decrease in primary production. Lower amounts of phytoplankton, in turn, reduce the population of algavorous ciliates [Jones 1992]. Size classes of ciliates in individual zones of the peatbog pools studied developed similarly. In the peat mat zone, in the transitional zone, and in the open water zone, species with sizes of 50–200 μm predominated in the majority of the peatbog pools studied. The rarest were ciliates with a size of less than 50 μm. Different results were obtained for a few peatbog pools in the Łęczna-Włodawa Lakeland, and for humic lakes, clearly predominated by small ciliate species [Carrick and Fahnenstiel 1990, Mieczan 2007a]. One of the features decisive of the distinctiveness of peatbog pool zones in terms of size classes of ciliates was the participation of the largest species of > 200 μm. In general, this class contributed more to the population in the peat mat zone. This may be related to lower pH values in this zone. Beaver and Crisman [1981], who observed a gradual increase in the participation of larger ciliates along with increasing acidity, came to similar conclusions.

CONCLUSIONS

In conclusion, the present study showed clear horizontal distribution patterns of ciliates. The highest abundance and biomass of protozoans were observed in the contact zone, while the lowest values were observed in the peat mat or open water zones. The results of this study suggest that the temperature, conductivity and total organic carbon are more important than concentrations of total phosphorus in limiting the horizontal distribution of planktonic ciliates.

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HORYZONTALNE ROZMIESZCZENIE ORZĘSKÓW POMIĘDZY PŁEM TORFOWCOWYM A STREFĄ OTWARTEJ WODY W PŁYTKICH TORFIANKACH

Streszczenie. Celem pracy była analiza zróżnicowania gatunkowego oraz liczebności orzęsków planktonowych w kilku torfiankach w układzie horyzontalnym: pło torfowcowe, strefa przejściowa – pło torfowcowe/woda oraz strefa otwartej wody. Ponadto analizowano wpływ wybranych właściwości fizycznych i chemicznych wód na występowanie tych mikroorganizmów. Zarówno bogactwo gatunkowe, jak i obfitość orzęsków były wyraźnie zróżnicowane w poszczególnych strefach. Największą różnorodność i liczebność orzęsków stwierdzono w strefie stykowej. Najbardziej uboga jakościowo i ilościowo okazała się natomiast strefa otwartej wody. Na stanowiskach zlokalizowanych wśród pła torfowcowego oraz w strefie stykowej największą liczebność osiągały bakteriożerne *Cinetochilum margaritaceum* oraz *Colpoda* sp. W strefie otwartej wody wzrastała natomiast liczebność mikotroficznego *Strombidium viride* oraz wszystkożernej *Vorticella companula*. Czynnikami w największym stopniu wpływającymi na występowanie protozooplanktonu w strefie pła torfowcowego oraz strefie stykowej były temperatura wody, przewodnictwo oraz zawartość całkowitego węgla organicznego. W strefie otwartej wody wzrastała natomiast liczba i siła powiązań pomiędzy obfitością orzęsków a zawartością w wodzie fosforu ogólnego.

Słowa kluczowe: torfowiska, torfianki, ekotony, torfowce, orzęski