

**24-HOUR CYCLE OF VARIABILITY IN CONTENTS
OF NITROGEN FORMS IN THE SURFACE MICROLAYER
OF THE BALTIC SEA LAGOON LAKE (NORTH POLAND) – PART I**

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Abstract

In July 2000 the diurnal cycle of analyses was conducted in order to determine variation in the concentrations of nitrogen and calcium compounds as well as dissolved gases in surface microlayers of a shallow coastal barrier lake located at the Baltic Sea coast. Water for analyses was collected in a diurnal cycle for the period of 24 h from lake Dołgie Wielkie. The analyses were conducted on the surface microlayer with a thickness of approx. 100 µm, a layer with a thickness of approx. 250 µm and a layer of subsurface water from a depth of 15 cm. Based on the analyzed data, information was collected on the migration of nitrogen compounds between the analyzed layers. Changes were observed in nitrogen forms reduced to oxidized forms and vice versa.

Key words: surface microlayer, lagoon lake, nitrogen, daily changes

INTRODUCTION

The analyzed surface microlayer is a thin layer found at the contact zone of the two environments: the hydrosphere and the atmosphere (Estep et al. 1985, Trojanowski et al. 2001). Knowledge on subsurface water microlayers is based on marine studies and relatively few studies are carried out in freshwater environments. This type of layers is formed at the surface of all aquatic ecosystems. The surface microlayer of water bodies is a unique chemical and physical environment, different from subsurface water. Among other factors, adhesion forces, formed as a result of molecular attraction at the boundary of the two media, i.e. hydrosphere and atmosphere, together with their surface tension forces, are responsible for the physical stability of the surface microlayer. This surface water layer is the boundary layer of extreme importance for both environments (Falkowska 1996, Hilbricht-Ilkowska et al. 1997). Exchange processes of gases and matter, variable in time and space, occur through

this layer (Falkowska 1996, Hilbricht-Ilkowska et al. 1997, Trojanowski et al. 2001). The exchange of substances and energy is a crucial process in the biogeochemical cycle of chemical elements, as it is the case in the aquatic environment. The surface microlayer affects gas exchange (Quinn and Otto 1971, Liss 1977) and transport mechanisms from the column of water to the atmosphere and *vice versa* (Norkrans 1980). Transport of substances occurs simultaneously between individual layers forming the surface microlayer. Dissolved substances, particles and microorganisms are transported to this zone by simple diffusion, convection by gas bubbles, convection movements from bottom deposits and subsurface water, as well as with migration organisms; moreover, the surface microlayer is fed by dusts and aerosols in precipitation (Norkrans 1980, Falkowska 1996). All these above mentioned processes lead to the accumulation of chemical substances and microorganisms.

Concentrations of various chemical and microbiological components found in this microlayer usually exceed their concentrations in subsurface water (Trojanowski et al. 2001). Among other things, markedly higher concentrations were found for heavy metals (Lion and Leckie 1981, Maki and Hermansson 1994), phosphorus and nitrogen compounds (Trojanowski et al. 2001, Mudryk et al. 2002, Falkowska 1999), sulphur compounds (Yang et al. 2005), fatty acids, esters, alcohols (Kozarec et al. 2003), proteins (Lion and Leckie 1981), chlorophyll (phytoplankton) (Hilbricht-Ilkowska et al. 1997, Falkowska 1999) as well as microorganisms (Maki and Remsen 1989, Mudryk et al. 2002, Hilbricht-Ilkowska and Kostrzewska-Szlakowska 2004).

Concentrations of nitrogen compounds found in the surface microlayer may undergo changes in the seasonal cycle, which is connected with the alternating seasons of the year (Antonowicz et al. 2008), as well as the diurnal cycle (Antonowicz 2008a, b). These fluctuations result from the activity of microorganisms responsible for these changes, e.g. redox changes and bioaccumulation, release, but also the above mentioned phenomena of transport of substances between water layers.

The aim of the study was to investigate the accumulation capacity of nitrogen forms, calcium and dissolved gases in relation to variation in concentrations of these substances throughout 24-hour variability in the surface microlayers of a lagoon lake situated in the Baltic coastal zone.

MATERIAL AND METHODS

Studied lake

A shallow and eutrophic lake Dołgie Wielkie (Fig. 1) is localized at 54°20.5' and 181°31.6'. The area of both basins in the lake is 156.4 ha, while the maximum depth is 2.9 m (Atlas jezior... 1997). The water table is located at 1.5 m a.s.l. The lake is situated in the Slovinski National Park (Poland).

Sampling

Samples of the surface water microlayer from the lake were collected over the period of 24 hours in June 2000. Sampling stations were established on the lake Dołgie Wielkie (Fig. 1) in a small bay. The selected sampling stations are shielded by trees

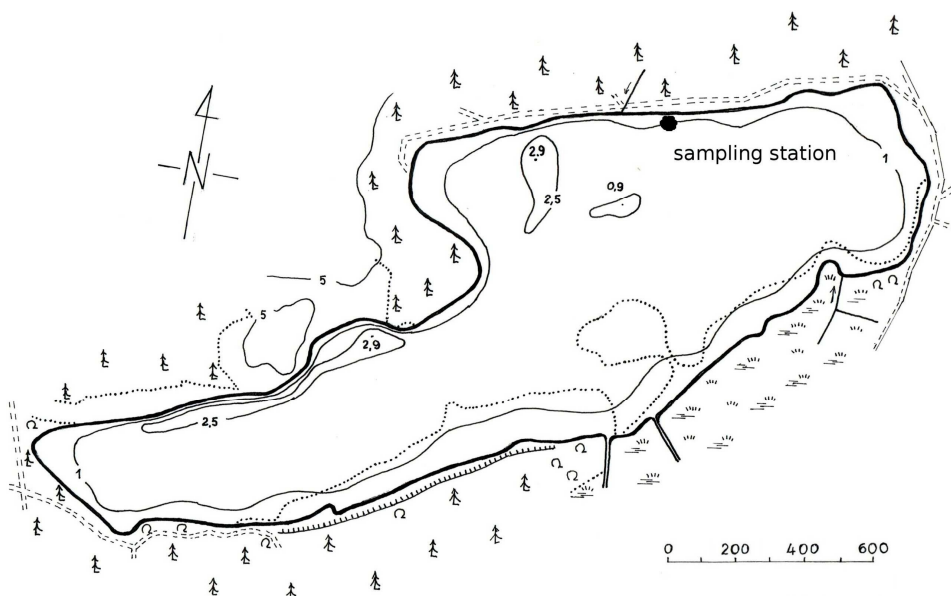


Fig. 1. Location of sampling stations on lake Dołgie Wielkie

growing at the lake shore, which reduces the effect of winds on the water surface at the sampling stations, and thus enhances the stability of the surface microlayer.

Samples of surface water were collected by three methods:

1. The plate method (PM) – (Harvey and Burzel 1972) was applied to the sample of surface water microlayer of 80-115 μm in thickness. For this purpose a glass plate of 30 x 30 cm and a thickness of 3 mm was used. A plate was immersed in water at an angle of 45° and then taken out at a rate of 5 to 20 $\text{cm}\cdot\text{s}^{-1}$. The water sample was removed from both sides of the plate to a container using a rubber wiper.
2. The Garrett method (SM) – (Garrett 1965) was used to collect the surface microlayer with a thickness of 250-300 μm . A polyethylene net of 75 x 75 cm and mesh size of 0.14 mm was mounted on a frame. The net was immersed in water in a similar manner as it was in case of the glass plate and next a wiper was used to collect water to a container (Garrett 1965, Piotrowicz et al. 1972). In order to avoid the dilution of the surface microlayer by the expansion of the material to the layer of subsurface water the boat used during sampling was allowed to float in the sampling area (Estep and Remsen 1985).
3. Subsurface water (SSW) was collected at a depth of about 15 cm from the water surface by immersing the container (Hermanowicz et al. 1999).

Chemical analyses

Total nitrogen according to Kjeldahl, organic nitrogen, ammonia nitrogen, carbon dioxide and dissolved oxygen were assayed according to Hermanowicz et al. (1999). Concentration of nitrite nitrogen was determined using a MERCK test.

Statistical analysis

In order to compare both investigated media, i.e. water from the surface microlayer and subsurface water, enrichment factors (EF) were applied, which were calculated from the following formula:

$$EF_{PM} = C_{PM}/C_{SSW}, EF_{SM} = C_{SM}/C_{SSW},$$

where: C_{PM} or C_{SM} – concentration of the analyzed component in a respective surface microlayer;

C_{SSW} – concentration of the same component in subsurface water (Estep et al. 1985, Guitart et al. 2004), counting in terms of the partial values:

$$EF = \frac{\sum \left(\frac{C_{SML1}}{C_{SSW1}} + \frac{C_{SML2}}{C_{SSW2}} + \dots + \frac{C_{SMLi}}{C_{SSWi}} \right)}{i},$$

where: successive unit indexes were calculated according to individual partial values, e.g. C_{SML1} – concentrations in season 1 in microlayer PM or SM, $_{SSW1}$ – concentrations of a component at the same hour and at the same sampling station in the subsurface water layer and analogously the 2nd results up to the i -th result.

When analyzing correlations of two variables Pearson's linear correlation coefficient r was calculated and next its significance was verified. If at least one distribution was significantly different from the normal distribution, the Spearman rank test r_s was applied. In order to determine the power of the relationship of correlated variables a scale presented by Stanisz (1998) was used. Based on the analysis of correlation, linear regression curves were selected and plotted.

RESULTS AND DISCUSSION

In the thinnest analyzed surface microlayer the highest concentrations of analyzed chemical substances were recorded, for nitrogen according to Kjeldahl it was a mean 1.01 mg dm^{-3} , that of organic nitrogen was by approx. 1% lower, the ammonia form of nitrogen was observed at $84.75 \text{ } \mu\text{g dm}^{-3}$, while the nitrite form accounts for $2.42 \text{ } \mu\text{g dm}^{-3}$ (Table 1). Calcium concentration was 9.80 mg dm^{-3} . In the GM layer the following mean values were observed: Kjeldahl nitrogen at 0.91 mg dm^{-3} , organic nitrogen at 0.90 mg dm^{-3} , ammonia nitrogen at $79.92 \text{ } \mu\text{g dm}^{-3}$, nitrite nitrogen at $2.33 \text{ } \mu\text{g dm}^{-3}$ and calcium at 9.22 mg dm^{-3} (Table 2). The lowest values were recorded in the subsurface water layer for each of the analyzed components (Table 3) and this result is consistent with those reported in studies by e.g. Trojanowski et al. (2001), Mudryk et al. (2002), Estep and Remsen (1985), Hillbricht-Ilkowska et al. (1997). In the neighbouring estuarine lake Gardno recorded concentrations of ammonia nitrogen in the surface microlayer were comparable (Trojanowski et al. 2001, Mudryk

Table 1
Contents of selected forms of nitrogen and calcium in surface microlayer
sampled by glass plate

SM	N-T	N _{org}	N-NH ₄	N-NO ₂	Ca
Min.	0.74	0.74	65.00	1.50	8.82
Med.	0.90	0.89	81.00	2.60	9.85
Max.	1.82	1.82	133.00	2.60	10.62
Mean	1.01	1.00	84.75	2.42	9.80
SD	0.28	0.29	14.64	0.42	0.50
SE	0.06	0.06	2.99	0.09	0.10
CV	28.09%	28.80%	17.27%	17.33%	5.10%

Min. – minimum, med. – median, max. – maximum, SD – standard deviation,
SE – standard error

Table 2
Contents of selected forms of nitrogen in surface microlayer sampled
by the Garrett net

GM	N-T	N _{org}	N-NH ₄	N-NO ₂	Ca
Min.	0.59	0.59	66.00	1.50	8.02
Med.	0.90	0.83	76.50	1.50	9.35
Max.	1.51	1.51	102.00	3.70	10.02
Mean	0.91	0.90	79.92	2.33	9.22
SD	0.23	0.23	10.58	0.99	0.57
SE	0.05	0.05	2.16	0.20	0.12
CV	25.16%	25.90%	13.24%	42.43%	6.18%

Table 3
Contents of selected forms of nitrogen, calcium and dissolved gases in surface microlayer
sampled by the Garrett net

PW	N-T	N _{org}	N-NH ₄	N-NO ₂	O ₂	CO ₂	Ca
Min.	0.53	0.53	61.00	1.50	5.00	2.50	8.42
Med.	0.76	0.76	76.00	1.50	9.80	3.90	9.22
Max.	0.95	0.95	99.00	3.70	12.9	5.10	9.42
Mean	0.77	0.77	78.75	2.05	9.22	3.93	9.11
SD	0.09	0.09	11.85	0.79	1.99	0.65	0.26
SE	0.02	0.02	2.42	0.16	0.41	0.13	0.05
CV	11.76%	11.76%	15.04%	38.76%	21.61%	16.60%	2.81%

Table 4
Enrichment coefficients calculated for analyzed substances

	N-T	N _{org}	N-NH ₄	N-NO ₂	Ca
EFSM	1.33	1.32	1.09	1.31	1.08
EFGM	1.20	1.18	1.03	1.21	1.01

et al. 2002). A significant value showing the difference in the concentrations of substances in the subsurface water layer and their concentrations in the surface microlayer is offered by enrichment factors presented in Table 4. It results from that table that each of the analyzed components enriched more the PM surface microlayer rather than SM microlayer. In the PM microlayer for nitrogen compounds there were values ranging from 1.09 for the reduced form of nitrogen (N-NH₄) to 1.33 for nitrogen according to Kjeldahl (N-T). In the SM layer located slightly deeper these coefficients ranged from 1.03 (N-NH₄) to 1.21 (N-NO₂). These values coincide with the lower threshold for enrichment factors recorded for N-NO₂ and N-NH₄ by Estep and Remsen (1985) in ponds, which may be considered comparable to the small lake of Dołgie Wielkie. For calcium this factor was the lowest and this means that the accumulation of this element in the surface microlayer was slight. This result is consistent with the findings from analyses conducted in three objects, i.e. Dołgie Wielkie, Jasień and Gardno, lakes located in the Polish Central Pomerania region (Antonowicz 2008c).

Concentrations of dissolved carbon dioxide and oxygen showed a negative significant correlation (Fig. 2). It is a result of oxygen uptake by microorganisms and release of carbon dioxide. This is confirmed by the analysis of graphs of diurnal concentrations of oxygen and carbon dioxide. In their case we can observe maximum concentrations of oxygen at time points of 10:00, 12:00, 17:00, 22:00, 2:00, 4:00 and 9:00 (the recorded time of sampling was approximated to the full hour), at which reduced values of carbon dioxide were found, and conversely – at time points, in which minimum oxygen concentrations were observed, maximum concentrations of dissolved carbon dioxide were recorded.

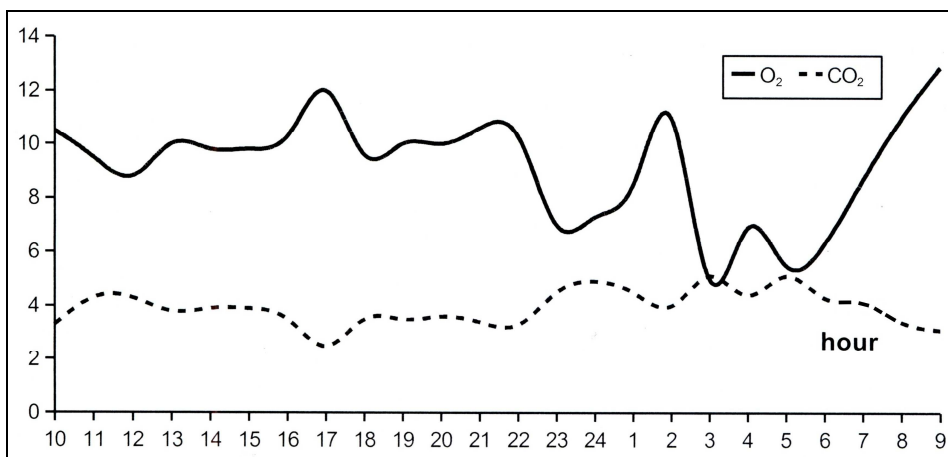


Fig. 2. Diurnal variation in the concentration of dissolved oxygen and carbon dioxide (mg dm⁻³) in subsurface water of lake Dołgie Wielkie

In case of the analyzed forms of nitrogen, calcium and dissolved gases a trend may be seen for similar concentrations in each of the investigated layers at the time of study onset and 24 h after that time, which suggests cyclicity of the observed changes.

Nitrogen according to Kjeldahl showed similar changes within the period of 24 h as those for organic nitrogen (which accounts for 99% N-T), which is markedly evident by the occurrence of common maxima in the surface microlayer observed at 22:00-24:00 (Figs 3 and 4). In that time frame in the subsurface water layer these forms of nitrogen showed a downward trend and were correlated with the decreasing level of oxygen (Fig. 2). This suggests that organic nitrogen forms migrate at that time to the microlayers. At the same time a decrease in oxygen concentration was accompanied by a decrease in the concentration of the nitrite form (Fig. 5) and the increase in the content of the reduced ammonia form N-NH_4 (Fig. 6) due to the enhanced reduction conditions. Observed maximum concentrations of total nitrogen (N-T and N_{org}) indicate an accumulation of organic matter, dying and decaying probably after 24:00, which in turn is connected with mineralization and the appearance of successively increasing amounts of the reduced forms of N-NH_4 and N-NO_2 in the PM microlayer, which

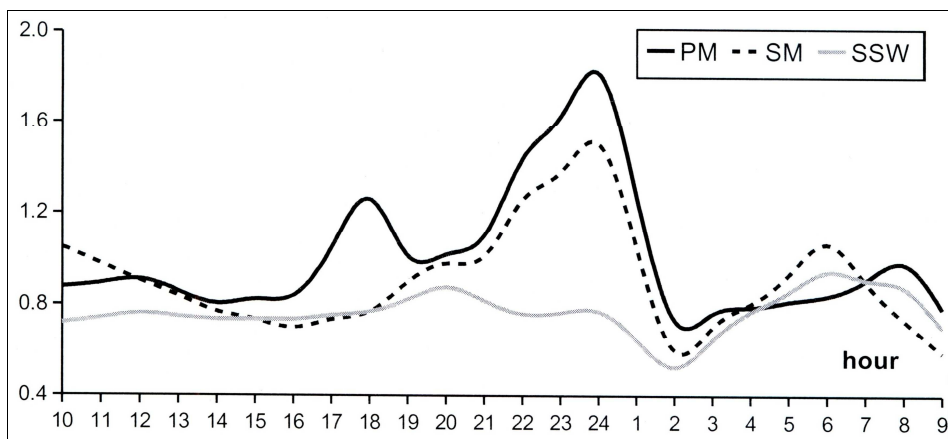


Fig. 3. Diurnal variation in the concentration of nitrogen according to Kjeldahl (mg dm^{-3}) in surface microlayers and subsurface water of lake Dołgie Wielkie

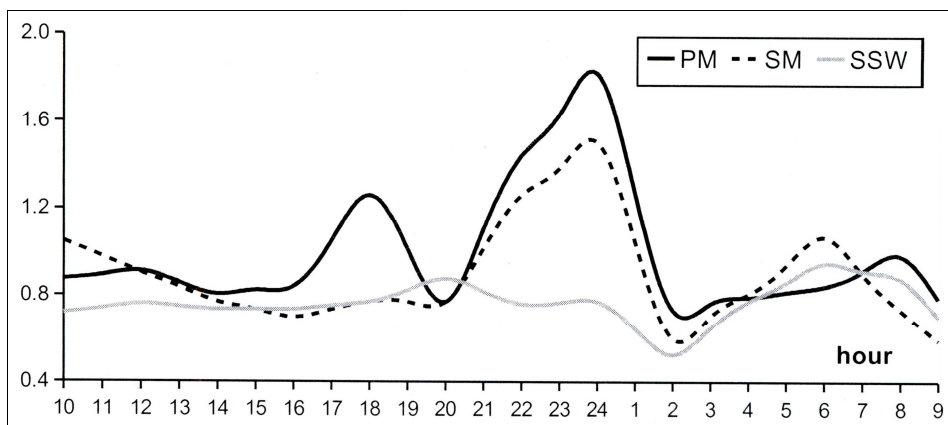


Fig. 4. Diurnal variation in the concentration of organic nitrogen (mg dm^{-3}) in surface microlayers and subsurface water of lake Dołgie Wielkie

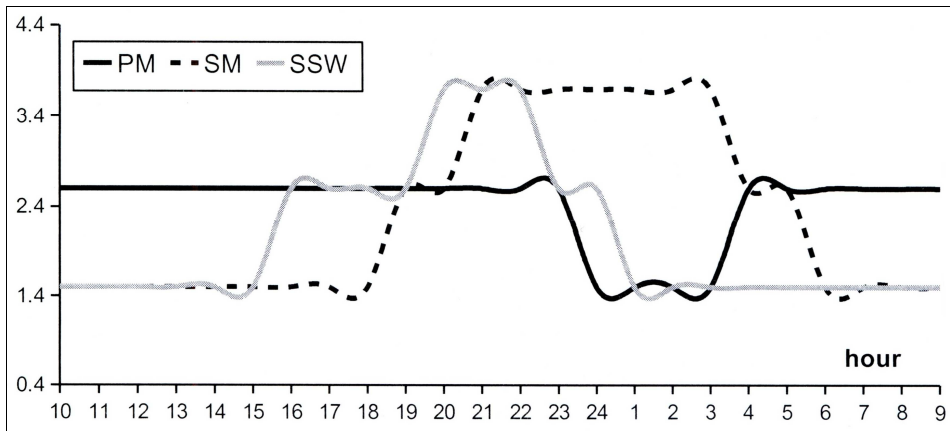


Fig. 5. Diurnal variation in the concentration of nitrite nitrogen ($\mu\text{g dm}^{-3}$) in surface microlayers and subsurface water of lake Dołgie Wielkie

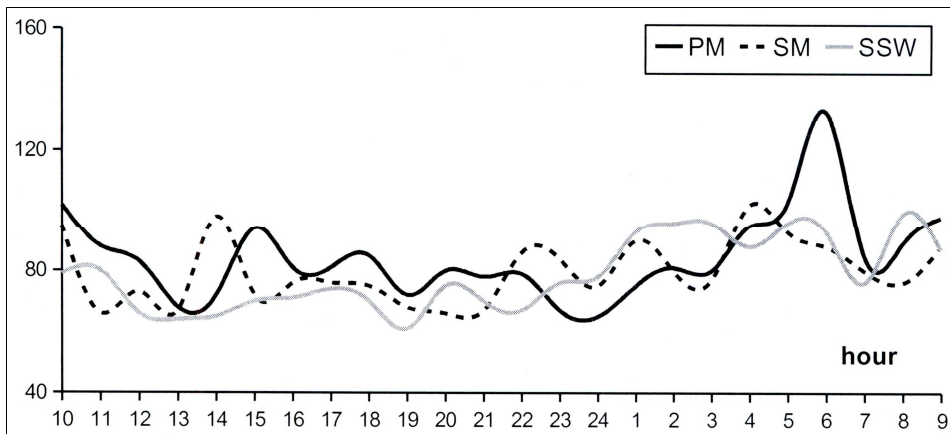


Fig. 6. Diurnal variation in the concentration of ammonia nitrogen ($\mu\text{g dm}^{-3}$) in surface microlayers and subsurface water of lake Dołgie Wielkie

could have been formed in the ammonification stage processes or possibly nitrification of the formed ammonia nitrogen. When analyzing this situation we need to focus on the low oxygen content in the subsurface water layer in the period of 3:00-6:00, which promotes reduction processes. This explains the simultaneous highest concentration in the PM microlayer recorded for the reduced form of N-NH_4 and a decrease in the N-NO_2 concentration. At that time considerable amounts of N-NO_2 migrate from microlayer PM to SM. Since in the SM microlayer a reduction is observed in the amounts of N-NH_4 , at the simultaneous increase in the concentration of this component in the PM microlayer and in the SSW layer, it may be assumed that the thicker microlayer was left by ammonia-forming microorganisms, which migrated to the water closer to the surface or to the depths, or in the SM layer the discussed ammonification occurred to a lesser degree, since at that time an increased concentration of organic matter was observed in the SM layer.

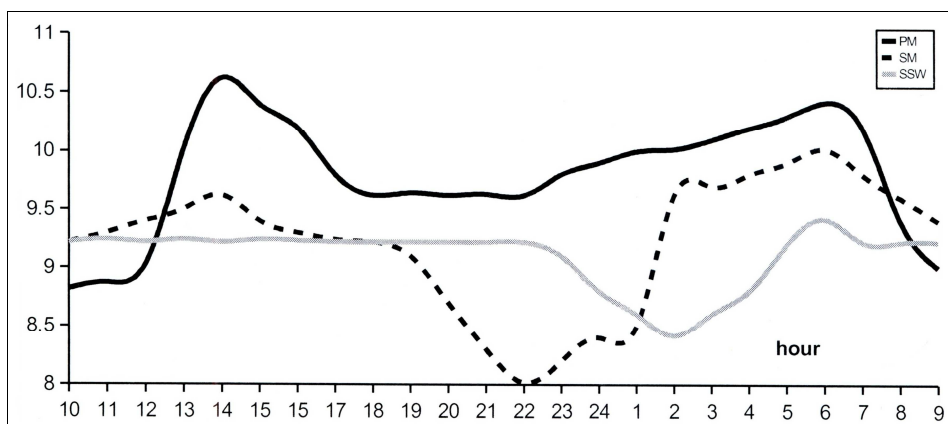


Fig. 7. Diurnal variation in the concentration of calcium (mg dm^{-3}) in surface microlayers and subsurface water of lake Dołgie Wielkie

When analyzing the diurnal variation in calcium concentration we need to stress the observed minimum (8 mg dm^{-3}) concentration of this element in layer SM at 22:00 (Fig. 7). In that time frame it was correlated with an increase in the concentration of organic matter (N_{org}) in that layer. Probably calcium is absorbed by organisms migrating to the depths of the water lake or it is precipitated in the form of sparingly soluble compounds. After 22:00 calcium concentration was reduced in the subsurface water layer and its marked increase was found in surface microlayers, i.e. calcium is transported in the opposite direction. At the same time we need to remember that an increase in calcium concentration starting from 1:00 a.m. is correlated with a decrease in the concentration of organic matter in the microlayers. Probably the limited capacity and the ability to accumulate substances in the surface microlayer led to a situation when calcium compounds displaced organic matter (N_{org}) from the surface microlayer to the SSW layer.

In literature there is a limited body of materials referring to diurnal variation. Scarce papers were published, some studies conducted in the marine environment (Falkowska 2001), while other conducted also in lakes of the Central Pomerania region, the latter concerning diurnal variation in the contents of metals, nutrients and phytoplankton (Antonowicz 2008a, b), as well as publications concerning bacterial activity (Walczak 2008).

Analysis of investigations conducted in October 2000 on the neighboring lake Gardno showed similarities in the course of diurnal variation in concentrations of total and organic nitrogen. Both in lake Gardno (Antonowicz 2008b) and lake Dołgie Wielkie in the diurnal cycle in the night hours maximum N-T concentration was observed in the surface microlayer. Moreover, in lake Gardno in the SM microlayer a maximum was recorded in the early morning hours (after 4:00). In case of ammonia nitrogen in both analyzed testing cycles maximum concentrations of ammonia nitrogen were found in the SM microlayer (lakes Gardno and Dołgie Wielkie). In the conducted investigations simultaneous maxima were observed on calcium concentrations found after 14:00 as well as at 4:00-5:00, similarly as in lake Gardno (Antonowicz 2008b) and the neighbouring lagune lake Dołgie Wielkie.

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24-GODZINNY CYKL ZMIENNOŚCI STĘŻENIA FORM AZOTU W MIKROWARSTWIE POWIERZCHNIOWEJ W LAGUNOWYM JEZIORZE MORZA BAŁTYCKIEGO (PÓŁNOCNA POLSKA) – CZĘŚĆ I

Streszczenie

W lipcu 2000 roku przeprowadzono dobowy cykl badań w celu określenia zmienności stężenia związków azotu oraz wapnia i rozpuszczonych gazów w mikrowarstwach powierzchniowych płytkiego jeziora lagunowego usytuowanego na wybrzeżu Morza Bałtyckiego. Wodę do analiz pobierano w cyklu godzinnym przez okres 24 godzin z jeziora Dołgie Wielkie. Poddano analizie mikrowarstwę powierzchniową o grubości ok. 100 μm , warstwę o grubości ok. 250 μm oraz warstwę wody podpowierzchniowej z głębokości około 15 cm. Na podstawie zanalizowanych danych uzyskano informacje dotyczące migracji związków azotu między analizowanymi warstwami. Zaobserwowano przemiany form azotu zredukowanych do utlenionych i *vice versa*.

