

## Diatom assemblages as indicators of salinity gradients: a case study from a coastal lake

by

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### Abstract

In order to understand the variability of diatoms in coastal lakes and its relationship to salinity, the authors have conducted a two-year study at Lake Resko Przymorskie (the Southern Baltic coast), which has a salinity between 1.9-4.8 PSU. Redundancy analysis (RDA) was used to describe the relationship between the species composition and selected variables. Four of the 10 measured variables of surface water chemistry (Cl<sup>-</sup> and PO<sub>4</sub><sup>3-</sup> concentrations, temperature, and pH) significantly explained 23% of the variation in the diatom species composition.

We found 82 taxa of diatoms (mostly tychoplanktonic) and determined the optimum and tolerance levels of salinity for predominant taxa (49 species with minimum 2% abundance). The optimum chloride concentration for the predominant diatoms ranged from 1471 to 2961 mg Cl<sup>-</sup> l<sup>-1</sup>.

The most abundant brackish water species was *Pseudostaurosira geocollegarum*. Brackish-freshwater diatoms were represented by *Cyclotella atomus*, *Cyclotella meneghiniana*, *Diatoma tenuis* and *Staurosira subsalina*. The most abundant fresh-brackish water diatoms were *Amphora pediculus*, *Fragilaria sopotensis*, *Hippodonta hungarica*, *Pseudostaurosira brevistriata* and *Staurosira construens*. Freshwater taxa accounted for as little as 1% of the population. This study provides new data on the ecology of coastal lakes and the possibility of using diatom-based transfer functions in the reconstruction of past environmental changes.

**Key words:** diatom, salinity preferences, transfer function, training set, environmental gradients, coastal lakes, Southern Baltic

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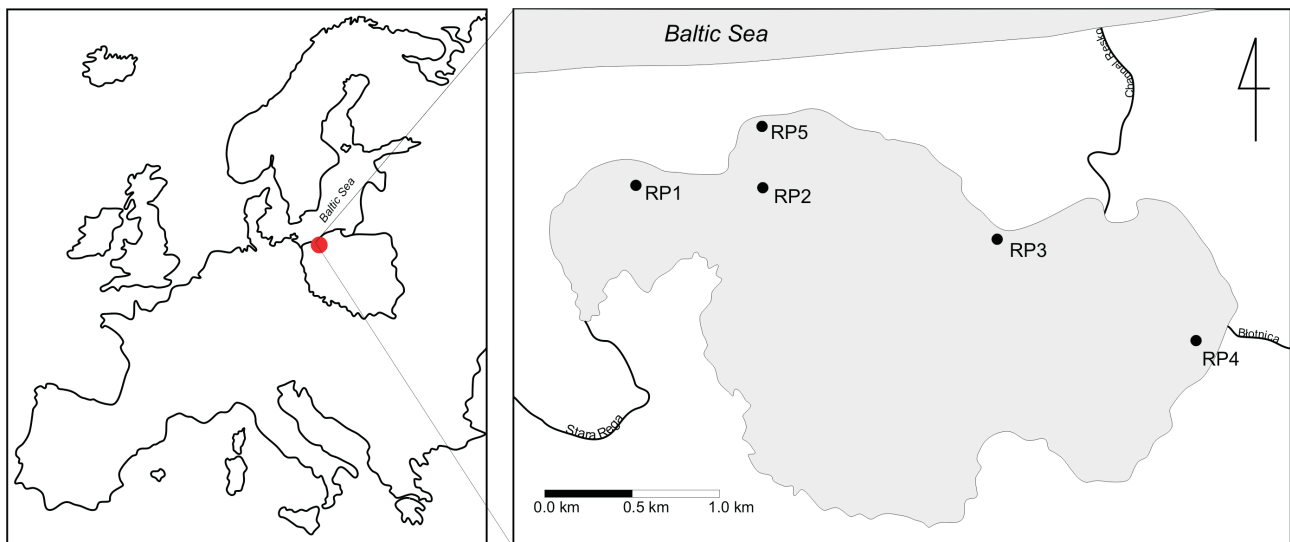
## Introduction

Diatoms (Bacillariophyceae) are a group of single-cell algae with cell walls saturated with silica, constituting approximately half of the cell dry mass. Many diatom species have well-defined ecological preferences and may therefore act as useful bioindicators. Diatoms are frequently encountered in waters as plankton, benthos and periphyton. In fresh waters, they account for up to 20-50% of the phytoplankton biomass, and their subfossil remains are very well preserved in sediments (Werner 1977; Battarbee 1986; Kawecka & Eloranta 1994; Stoermer & Smol 1999; Battarbee et al. 2001). They are mostly used in the assessment of water quality in modern water ecosystems (Reid et al. 1995; Kelly et al. 2009; Kelly & Zgrundo 2013), and in the reconstruction of past transformations of water bodies (Battarbee et al. 1999; Fritz et al. 1999; Zong & Horton 1999; Battarbee et al. 2001; Zong & Sawai 2015). Modern palaeoenvironmental reconstructions based on the analysis of fossil diatom assemblages extensively employ the so-called transfer functions, which quantitatively determine the correlations between the occurrence of taxa and environmental variables (Battarbee & Charles 1986; Fritz et al. 1991; Sweets 1992; Ng & King 1999; Westman & Sohlenius 1999; Jones & Birks 2004; Paul et al. 2010).

Current studies show that the abundance and species structure of diatoms are controlled by a number of environmental variables. Water pH, trophic conditions, and temperature have often been identified among the important factors determining diatom assemblages in fresh waters

(Żelazna-Wieczorek 2011; Hildebrandt-Radke et al. 2012; Kittel et al. 2014; Petera-Zganiacz et al. 2015; Fiłoc et al. 2016). Diatoms are also known to respond strongly to salinity (Juggins 1992; Cumming & Smol 1993; Fritz et al. 1993; Saunders 2011; Zong & Sawai 2015). The latter parameter represents one of the most important environmental variables in coastal lakes that have experienced spatial and temporal changes in salinity when sea levels increased prehistorically as well as more recently. Research on long-term changes in salinity in coastal lakes elucidates changes in sea levels over the last centuries and millennia. Diatom analysis is one of the primary tools in the research on changes in salinity of seas, lagoons and estuaries in various geographical regions (Gillieson 1991; Douglas et al. 1996; Roberts & McMinn 1999; Taffs 2001; Wachnicka et al. 2010; Saunders 2011; Majewska et al. 2012).

In the area of the Polish coast, diatom analyses were performed for sediments from lakes: Jamno (Przybyłowska-Lange 1979; Miotk-Szpiganowicz et al. 2007; Miotk-Szpiganowicz et al. 2008), Dołgie Wielkie (Mazurek et al. 2008; Lutyńska 2008a), Sarbsko (Przybyłowska-Lange 1981; Miotk & Bogaczewicz-Adamczak 1986; Miotk-Szpiganowicz et al. 2007; Miotk-Szpiganowicz et al. 2008; Woszczyk et al. 2008; Woszczyk et al. 2010; Woszczyk et al. 2014), Gardno (Bogaczewicz-Adamczak 1977; Bogaczewicz-Adamczak et al. 1981; Bogaczewicz-Adamczak & Miotk 1985; Miotk-Szpiganowicz et al. 2007; Lutyńska 2008b; Lutyńska 2008c; Miotk-Szpiganowicz et al. 2008; Lutyńska & Rotnicki 2009) and Łebsko (Staszak-Piekarska & Rzodkiewicz 2015). The results of these



**Figure 1**

Location of study sites. The inset shows location of Lake Resko Przymorskie

analyses describe the development and functioning of coastal lakes during and after incursions of seawater during the last marine transgression (Rotnicki et al. 1999; Rotnicki 2003; Woszczyk & Rotnicki 2009; Cedro et al. 2012).

The objectives of the present work were: (1) to determine the composition and structure of diatom assemblages of Lake Resko Przymorskie, (2) to identify the most important environmental parameters affecting diatom assemblages, and (3) to estimate salinity optima and tolerance of dominant diatoms.

## Study area

Lake Resko Przymorskie (15°22'37"E, 54°08'32"N) occupies the lowest part of the Rega and Błotnica River valleys (Dobracka 1992). The average long-term elevation of the water level is 2.2 m a.s.l., but the bottom of the lake is located in a cryptodepression. The lake is separated from the Baltic Sea by a dune embankment extending from Dźwirzyno to Mrzeżyno, with a maximum height of 17 m a.s.l. The surface area of the lake is 559.0 ha, the maximum depth 2.5 m, and the mean depth 1.3 m.

The area south of the lake was deforested in the 19<sup>th</sup> century and is largely under the agricultural use. The northern shore together with the entire spit separating the lake from the Baltic Sea remains almost entirely forested. Lake Resko Przymorskie is fed by several small streams, including the Łuzanka, the Bagienna, and the Stara Rega. The lake is connected with the sea in the northeastern part through the wide Resko Canal (Regoujście), which is almost one kilometer long. The free flow of water through the canal is possible owing to the absence of any obstacles. Therefore, saline water from the sea easily flows into the lake.

## Materials and methods

A total of 21 water samples were collected. Each sample was divided in two subsamples, one for diatoms and the other for water chemistry. Water samples were collected from the near-surface layer (0.5 m depth), between September 2012 and July 2014 (Fig. 1). Concentrations of chlorides (Cl<sup>-</sup>), phosphorus (PO<sub>4</sub><sup>3-</sup> and total phosphorus, P<sub>tot</sub>), and forms of nitrogen (NH<sub>4</sub><sup>+</sup> and NO<sub>3</sub><sup>-</sup>) were measured in the laboratory.

Water parameters, including salinity, pH, temperature, and dissolved oxygen (DO) were recorded during each sampling event using the HI9828 multiparameter meter, Hanna Instruments (Table 1).

**Table 1**

Classification merging salinity and chlorides content used in this study (after Van Dam et al. 1994)

Class	Description	Cl <sup>-</sup> (mg l <sup>-1</sup> )	Salinity (PSU)
1	fresh	< 100	< 0.2
2	fresh-brackish	< 500	< 0.9
3	brackish-fresh	500-1000	0.9-1.8
4	brackish	1000-5000	1.8-9.0

Before determining the ion components, water samples were filtered through Whatman's filter paper. Concentrations of NH<sub>4</sub><sup>+</sup> were determined using the spectrophotometric method (Spekol 1100, Carl Zeiss Technology) with Nessler's reagent. The levels of P<sub>tot</sub> and PO<sub>4</sub><sup>3-</sup> were determined by the spectrophotometric molybdate method with stannous chloride, following methods of Prejzner 1994 and Elbanowska et al. 1999. To determine total phosphorus (P<sub>tot</sub>), HNO<sub>3</sub> (1:1) and H<sub>2</sub>O<sub>2</sub> (6%) were added in the amount of 0.25 cm<sup>3</sup> per 50 cm<sup>3</sup> of sample, which was followed by mineralization in a UV mineralizer before spectrophotometry. Chloride and nitrate levels were determined with ion chromatography (ion chromatograph DX-120, Dionex). In the case of samples with high chloride content, the analysis was performed after appropriate dissolution of a sample.

## Diatom analysis

The diatom analysis was conducted according to the method of Battarbee (1986). Samples were treated with 10% HCl to remove calcium carbonate and washed several times in distilled water. Afterward, the samples were boiled in 30% H<sub>2</sub>O<sub>2</sub> in order to oxidize the organic matter. Permanent microscope slides were mounted in Naphrax. The identification of diatoms was performed using a Nikon Eclipse E-200 light microscope at a magnification of ×1000. At least 500 frustules of diatoms were counted on each slide and the relative abundance of each taxon in a sample was calculated. The diatoms were identified according to Krammer & Lange-Bertalot (1986, 1988, 1991a,b), Denys (1991/2), van Dam et al. (1994), Snoeijs (1993), Snoeijs & Vilbaste (1994), Snoeijs & Potapova (1995), Lange-Bertalot & Metzeltin (1996), Snoeijs & Kasprovičienė (1996), Snoeijs & Balashova (1998), Lange-Bertalot (1999), Lange-Bertalot & Genkal (1999), Krammer (2002), Lange-Bertalot et al. (2003), Hoffmann et al. (2011), Lange-Bertalot (2011), and Zgrundo et al. (2013). Data from AlgaeBase (Guiry & Guiry 2016) were used for the more recent diatom nomenclature.

The classification of diatoms, taking into consideration their ecological requirements (salinity, trophic state, pH), was based on the OMNIDIA database (Lecointe et al. 1993).

In terms of lifeforms, diatom taxa were grouped according to Denys' classification (1991/2), focusing mainly on euplanktonic and tychoplanktonic species (including tychoplanktonic, of eponitic origin and benthic origin). Salinity preferences of diatoms were based on the classification by van Dam et. al (1994; after van der Werff & Huls, 1954-1974; Table 1). In terms of the trophic spectrum, diatoms were grouped on the basis of Naumann's classification (1932), and pH classification – according to Hustedt (1939; see Denys 1991/2).

### Numerical analysis

The redundancy analysis (RDA) was used to determine similarities between the species distributions and sampling sites with respect to physical and chemical water parameters: salinity, temperature, pH, dissolved oxygen (DO), nitrogen (ammonium  $[\text{NH}_4^+]$ , and nitrate  $[\text{NO}_3^-]$ ), phosphorus (phosphate  $[\text{PO}_4^{3-}]$  and total phosphorous  $[\text{P}_{\text{tot}}]$ ) and chloride ( $\text{Cl}^-$ ). Diatoms with relative abundance above 2% were included in the analysis (49 species). RDA analysis was performed for all sites throughout the sampling period using the CANOCO 4.5 software (ter Braak & Šmilauer 2003). Furthermore, Monte Carlo permutation tests were run to reduce the number of variables (Lepš & Šmilauer 2003). All variables with a significance level of  $p > 0.05$  were removed. Chloride optima and tolerance ranges for diatom species were obtained using the C2 software (Juggins 2007), including the model weighted averaging (WA).

## Results

### Water chemistry

Most sites studied from Lake Resko Przymorskie were rich in nutrients, especially  $\text{NH}_4^+$  (mean 0.3-1.1  $\text{mg l}^{-1}$ ) and  $\text{PO}_4^{3-}$  (mean 0.05-0.1  $\text{mg l}^{-1}$ ), and had high  $\text{Cl}^-$  concentration (mean 1401-3160  $\text{mg l}^{-1}$ ). High  $\text{Cl}^-$  concentration is a significant habitat feature of this coastal lake. During the sampling period, water temperature varied from 3.2°C to 24.4°C (average 16.5°C). Table 2 presents the summary of hydrochemical features of all sites.

Individual study sites in Lake Resko Przymorskie showed a wide range of nutrient and chloride concentrations. Ammonium ( $\text{NH}_4^+$ ) occurred throughout the measurement period. Its mean concentration was 0.5  $\text{mg l}^{-1}$ . The lowest ammonium concentration was observed in May 2013 and July 2013, and the highest ammonium concentration was observed in July 2014.

Increased nitrate concentration ( $\text{NO}_3^-$ ) was observed only in samples from May 2013. The average concentration of phosphates ( $\text{PO}_4^{3-}$ ) was very low and varied from 0.05  $\text{mg l}^{-1}$  to 0.1  $\text{mg l}^{-1}$ . In July 2013 and May 2014, phosphates were not recorded. The mean concentration of total phosphorous ( $\text{P}_{\text{tot}}$ ) was 58  $\mu\text{g l}^{-1}$ . The lowest concentration was recorded in May 2013 (RP5), and the highest in September 2013 (sample RP4). The chloride concentrations during the study period showed considerable temporal variability. The highest values were determined in November 2013 (sample RP4, and sample RP5), and the lowest in May 2013 (sample RP4). The highest salinity of waters in Lake Resko Przymorskie was observed in November 2013 and the lowest in May 2013. The

**Table 2**

Average, maximum and minimum values of water quality variables recorded during sampling events

Season	Temp (°C)	Salinity (PSU)	DO (ppm)	pH	$\text{NH}_4^+$ ( $\text{mg l}^{-1}$ )	$\text{NO}_3^-$ ( $\text{mg l}^{-1}$ )	$\text{PO}_4^{3-}$ ( $\text{mg l}^{-1}$ )	$\text{P}_{\text{tot}}$ ( $\mu\text{g l}^{-1}$ )	$\text{Cl}^-$ ( $\text{mg l}^{-1}$ )
09.2012	14.9 (14.4 - 15.3)	3.5 (3.0-3.7)	10.4 (6.6-12.4)	8.4 (7.7-8.7)	0.6 (0.5-0.7)	n.d.	0.05 (0.0-0.1)	55.4 (42.0-78.0)	2495 (2049-2686)
05.2013	17.8 (17.0 - 19.2)	1.9 (1.9-2.0)	n.a.	9.0 (8.9-9.1)	0.3 (0.3-0.4)	2.5 (2.0-2.9)	0.1 (0.1-0.1)	42.3 (36.0-52.0)	1401 (1328-1451)
07.2013	21.3 (21.0-21.4)	2.5 (2.4-2.5)	9.4 (8.8-10.5)	9.1 (9.0-9.2)	0.3 (0.3-0.3)	n.d.	n.d.	67.8 (55.0-82.0)	2158 (2123-2195)
09.2013	14.2 (14.1-14.2)	3.4 (3.3-3.5)	11.1 (10.2-12.4)	9.2 (9.2-9.3)	0.6 (0.5-0.7)	n.d.	0.1 (0.1-0.1)	76.0 (65.0-85.0)	2687 (2541-2812)
11.2013	3.2 (3.2-3.2)	4.8 (4.7-4.8)	13.0 (12.1-13.9)	9.9 (9.8-9.9)	0.4 (0.3-0.4)	n.d.	0.1 (0.1-0.1)	35.0 (34.0-36.0)	3160 (3090-3231)
05.2014	21.3 (21.3-21.3)	3.6 (3.6-3.6)	11.3 (11.3-11.3)	8.5 (8.5-8.5)	0.6 (0.6-0.6)	n.d.	n.d.	31.0 (31.0-31.0)	2208 (2208-2208)
07.2014	24.1 (23.7-24.4)	3.1 (3.0-3.1)	8.2 (7.2-9.1)	9.5 (9.4-9.5)	1.1 (1.0-1.1)	n.d.	0.1 (0.1-0.1)	65.0 (54.0-76.0)	2306 (2234-2378)

n.a. - not analysed; n.d. - not detected

oxygenation of near-surface waters (DO) varied from 6.6 ppm in September 2012 (sample RP5) to 13.9 ppm in November 2013 (sample RP4). Water pH was rather high and ranged from 7.7 in September 2012 (sample RP5) to 9.9 in November 2013 (sample RP4).

**Diatom assemblages**

A total of 82 diatom taxa were identified in the analyzed samples. Tychoplanktonic (random planktonic) forms dominated (Fig. 2). From September 2012 to July 2014, the contribution of tychoplanktonic species (e.g., *Pseudostaurosira geocollegarum*, *Staurosira construens* and *Pseudostaurosira brevistriata*) was relatively high (from 72.6% to 99.0% average abundance). The abundance of typical planktonic (euplanktonic) species was rather low, from 0.8% to 24.1% average abundance in all samples. An increased contribution (mainly *Cyclotella atomus* and *Cyclotella meneghiniana*) was observed in July 2013 (Fig. 3).

The diatom assemblages were dominated by species with brackish, brackish-fresh and fresh-brackish salinity preferences. In September 2012, May 2013, and September 2013, brackish diatoms dominated. Their average contribution amounted to over 55%. In subsequent measurement seasons, the contribution of brackish taxa amounted to 34.1% average abundance in July 2013, 38.0% in November 2013, and more than 40.0% in May and July 2014. The dominant brackish species was *Pseudostaurosira geocollegarum*. Brackish-fresh

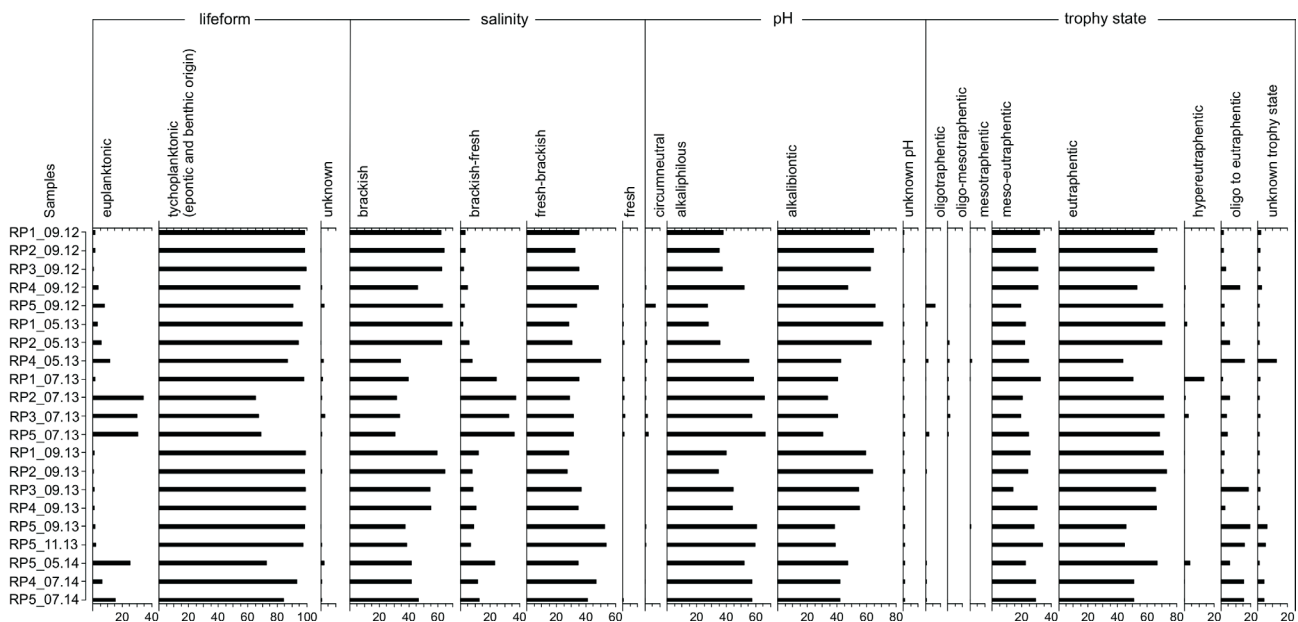
diatoms were most abundant in July 2013 (26.8% average abundance) and May 2014 (23.2% average abundance). They were the uncommon in September 2012 (3.1%) and May 2013 (5.1%). Brackish-fresh species in Lake Resko Przymorskie included: *Cyclotella atomus*, *Cyclotella meneghiniana*, *Pseudostaurosira subsalina* and *Diatoma tenuis*. The highest number of fresh-brackish taxa occurred in November 2013 (53.2% average abundance). In the remaining research seasons, their contribution amounted to more than 32.0%. Fresh-brackish species included: *Amphora pediculus*, *Fragilaria sopotensis*, *Hippodonta hungarica*, *Pseudostaurosira brevistriata* and *Staurosira construens*. Freshwater diatoms accounted for less than 1% of the assemblage.

The pH spectrum was dominated by alkalibiontic (from 37.7% to 60.6% average abundance) and alkaliphilous (from 37.8% to 60.4% average abundance) taxa. Diatoms with neutral water pH preferences were considerably less abundant (0.0-1.5% average abundance).

Among all taxa with the determined trophic preferences, eutraperthentic taxa were the most abundant group (from 45.3% to 69.0% average abundance). Eutraperthentic taxa were represented by *Pseudostaurosira brevistriata* and *Staurosira construens*.

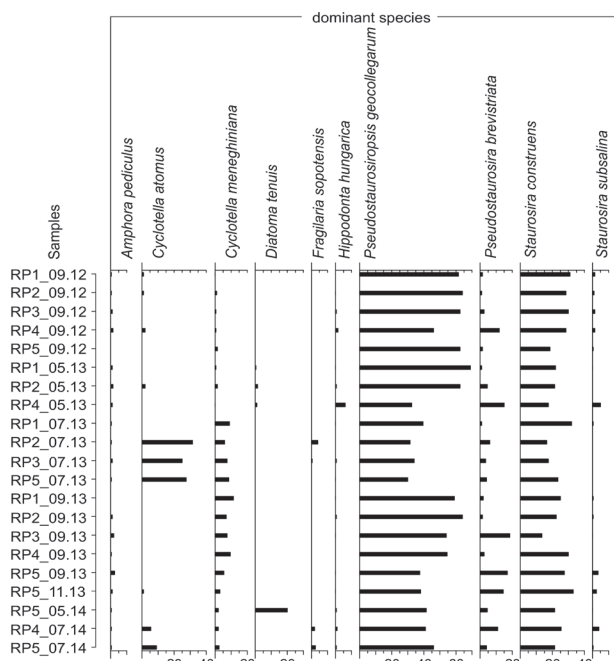
**Species-environment relationships**

The results of RDA analysis reflecting correlations between the frequencies of predominant diatoms and



**Figure 2**

Relative abundance of ecological groups of diatoms from September 2012 to July 2014



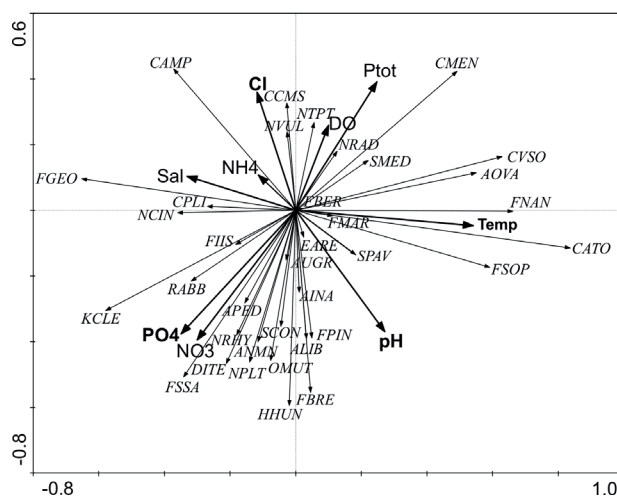
**Figure 3**  
Relative abundance of dominant species in Lake Resko Przymorskie (species with values >2%)

physicochemical water parameters are shown in Fig. 4. In the Monte Carlo test, 4 out of 10 environmental parameters appeared significant (Table 3), including: water temperature, pH,  $\text{PO}_4^{3-}$  and  $\text{Cl}^-$  concentrations. These 4 parameters together accounted for 23% of the variance in the diatom diversity. Temperature is particularly positively correlated with the first RDA axis, and chloride concentrations with the second axis.

RDA analysis revealed environmental requirements of different ecological groups. Figure 4 illustrates that the occurrence of *Pseudostaurosira geocollegarum* (the most abundant species), *Navicula cincta* and *Cocconeis placentula* var. *lineata* was correlated with higher salinity (Sal), while *Navicula vulpina* and *Cyclotella comensis* were correlated with high chloride content. *Caloneis amphisbaena* showed a high correlation with  $\text{NH}_4^+$ , and relatively low pH values. The occurrence of *Navicula tripunctata*, *Navicula radiosa*, *Cyclotella meneghiniana* and *Stephanodiscus medius* was related to the abundance of phosphorus and well-oxygenated waters ( $\text{P}_{\text{tot}}$ , DO). The occurrence of *Amphora ovalis*, *Cavinula scutelloides*, *Cyclotella atomus*, *Belonastrum berolinensis*, *Fragilaria nanana*, *Fragilaria sopotensis*, *Martyana martyi* and *Stephanodiscus parvus* was strongly determined by high water temperature. *Staurosirella pinnata*, *Amphora libyca*, *Amphora inariensis*, *Aulacoseira granulata* and *Ellerbeckia arenaria* showed a strong

correlation with pH. The occurrence of *Fragilaria inflata* var. *istvanffy*, *Rhoicosphenia abbreviata*, *Actinocyclus normanii*, *Amphora pediculus*, *Navicula rhynchocephala*, *Diatoma tenuis*, *Staurosira construens*, *Pseudostaurosira brevistriata*, *Pseudostaurosira subsalina*, *Placoneis placentula*, *Opephora olsenii* and *Hippodonta hungarica* was related to high concentrations of  $\text{NO}_3^-$  and a phosphate-rich environment.

The strongest correlation with salinity ( $\text{Cl}^-$ , salinity) occurred for samples from September 2012 (RP1, RP2,



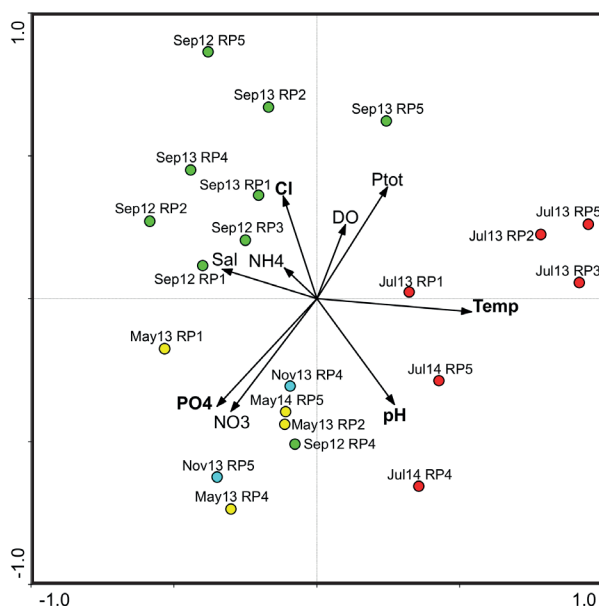
**Figure 4**  
Biplots of redundancy analysis (RDA): chemical and physical variables (Temp,  $\text{NO}_3^-$ ,  $\text{NH}_4^+$ ,  $\text{P}_{\text{tot}}$ ,  $\text{PO}_4^{3-}$ , pH,  $\text{Cl}^-$ , salinity, DO) and diatom species (species with values > 2% of relative abundance). Statistically significant variables are marked in bold. Code used for species: ANMN – *Actinocyclus normanii*, AINA – *Amphora inariensis*, ALIB – *Amphora libyca*, AOVA – *Amphora ovalis*, APED – *Amphora pediculus*, AUGR – *Aulacoseira granulata*, CAMP – *Caloneis amphisbaena*, CATO – *Cyclotella atomus*, CCMS – *Cyclotella comensis*, CMEN – *Cyclotella meneghiniana*, CPLI – *Cocconeis placentula* var. *lineata*, CVSO – *Cavinula scutelloides*, DITE – *Diatoma tenuis*, EARE – *Ellerbeckia arenaria*, FBER – *Belonastrum berolinensis*, FBRE – *Pseudostaurosira brevistriata*, FGEO – *Pseudostaurosira geocollegarum*, FIIS – *Fragilaria inflata* var. *istvanffy*, FMAR – *Martyana martyi*, FNAN – *Fragilaria nanana*, FPIN – *Staurosirella pinnata*, FSOP – *Fragilaria sopotensis*, FSSA – *Pseudostaurosira subsalina*, HHUN – *Hippodonta hungarica*, KCLE – *Karayevia clevei*, NCIN – *Navicula cincta*, NPLT – *Placoneis placentula*, NRAD – *Navicula radiosa*, NRHY – *Navicula rhynchocephala*, NTPT – *Navicula tripunctata*, NVUL – *Navicula vulpina*, OMUT – *Opephora olsenii*, RAAB – *Rhoicosphenia abbreviata*, SCON – *Staurosira construens*, SMED – *Stephanodiscus medius*, SPAV – *Stephanodiscus parvus*

**Table 3**

Results of the forward selection of environmental parameters (Monte Carlo permutation test in RDA,  $p < 0.05$  are statistically significant and given in bold)

Variable	$\lambda$	$F$	$p$
Temperature	0.11	2.39	<b>0.032</b>
pH	0.12	2.70	<b>0.015</b>
$PO_4^{3-}$	0.08	2.06	<b>0.047</b>
Salinity	0.06	1.46	0.155
$Cl^-$	0.08	2.09	<b>0.037</b>
DO	0.04	1.18	0.308
$P_{tot}$	0.04	1.16	0.295
$NO_3^-$	0.03	0.92	0.490
$NH_4^+$	0.03	0.98	0.434

RP3, RP5) and September 2013 (RP1, RP2, RP4) (Fig. 5), but sample RP5 from September 2013 was correlated with high values of total phosphorus ( $P_{tot}$ ). Summer samples from July 2013 (RP1, RP2, RP3, RP5) and July

**Figure 5**

Samples with respect to the chemical and physical water parameters (Temp,  $NO_3^-$ ,  $NH_4^+$ ,  $P_{tot}$ ,  $PO_4^{3-}$ , pH,  $Cl^-$ , salinity, DO). Statistically significant variables are marked in bold.

2014 (RP4, RP5) showed the strongest correlations with temperature, although in July 2014, pH also had a considerable impact (in addition to temperature). The strongest correlations with phosphates ( $PO_4^{3-}$ ) and nitrate ions ( $NO_3^-$ ) were found in September 2012 (sample RP4), May 2013 (samples RP1, RP2, and RP4), November 2013 (samples RP4 and RP5) and May 2014 (sample RP5).

### Optima and Tolerance of Diatom Species

We determined salinity preferences ( $Cl^-$  concentration) for 49 diatom species. The optimum chloride concentration for the identified diatoms ranged from 1471 mg  $Cl^- l^{-1}$  to 2961 mg  $Cl^- l^{-1}$  (Table 4). Thirty-nine taxa showed an optimum chloride concentration above 2000 mg  $l^{-1}$ . Four of the species (*Epithemia turgida*, *Stausosira binodis*, *Encymoena silesiacum* and *Amphora marina*) showed particularly high optima, exceeding 2600 mg  $Cl^- l^{-1}$ . Moreover, *E. turgida* and *S. binodis* were distinguished by narrow tolerance ranges (*E. turgida* had an optimum of 2961 mg  $Cl^- l^{-1}$  with a tolerance of 385 mg  $Cl^- l^{-1}$ , while *S. binodis* had an optimum of 2749 mg  $Cl^- l^{-1}$  with a tolerance of 324 mg  $Cl^- l^{-1}$ ). Particularly high tolerance to chloride concentration was revealed for the following species: *A. marina* (1159 mg  $Cl^- l^{-1}$ ), *Fragilaria capucina* var. *mesolepta* (1159 mg  $Cl^- l^{-1}$ ), *Planothidium lanceolatum* (1015 mg  $Cl^- l^{-1}$ ) and *Placoneis placentula* (964 mg  $Cl^- l^{-1}$ ). Species with optima below 2000 mg  $Cl^- l^{-1}$  belonged to 10 taxa. *Parlibellus crucicula* and *Planothidium delicatulum* were found to be indicator species for very low salinity (below 1600 mg  $Cl^- l^{-1}$ ) within the range of Lake Resko Przymorskie (Table 4).

## Discussion

### Gradients and Environmental Variables

There are many studies devoted to the general ecology of diatoms in salt waters in Poland (e.g. Andrén 1999; Bogaczewicz-Adamczak et al. 2001; Bogaczewicz-Adamczak & Dziengo 2003), however, in-depth research into relationships between populations of diatoms and various environmental variables is scarce (Zgrundo & Bogaczewicz-Adamczak 2004; Dziengo-Czaja et al. 2008). Diatoms found in Polish coastal lakes affected by saline waters are an important component of phytoplankton, though their ecology has not yet been carefully studied. What is more, relatively little is known about seasonal changes in diatom composition in coastal lakes. The influence of saline waters and salinity gradients are of crucial

Table 4

Optimum (weighted average) and tolerance (SD) of particular species compared to Cl<sup>-</sup> gradient

Species	Abundance			Cl <sup>-</sup> mg l <sup>-1</sup>	
	frequency	mean	SD	Optimum	Tolerance
<i>Epithemia turgida</i> (Ehrenberg) Kützing	2	0.0	0.1	2960.9	384.9
<i>Stausosira binodis</i> (Ehrenberg) Lange-Bertalot	2	0.0	0.1	2749.1	324.2
<i>Encymoena silesiacum</i> (Bleisch) D.G.Mann	2	0.0	0.1	2648.7	835.2
<i>Amphora marina</i> W.Smith	2	0.0	0.1	2638.9	1159.3
<i>Fragilaria capucina</i> var. <i>mesolepta</i> (Rabenhorst) Rabenhorst	2	0.0	0.1	2495.1	1159.3
<i>Navicula tripunctata</i> (O.F.Müller) Bory	3	0.1	0.2	2473.7	380.2
<i>Amphora ovalis</i> (Kützing) Kützing	3	0.1	0.1	2472.6	505.7
<i>Amphora pediculus</i> (Kützing) Grunow ex A.Schmidt	12	0.7	0.6	2459.1	618.3
<i>Cyclotella meneghiniana</i> Kützing	12	4.5	3.8	2457.3	391.5
<i>Pseudostaurosira brevistriata</i> (Grunow) D.M.Williams & Round	11	6.3	5.9	2414.3	630.2
<i>Staurosirella pinnata</i> (Ehrenberg) D.M. Williams & Round	4	0.5	1.0	2386.1	535.0
<i>Staurosira construens</i> Ehrenberg	20	24.5	6.1	2385.6	497.9
<i>Caloneis amphisbaena</i> (Bory) Cleve	5	0.1	0.1	2379.5	466.5
<i>Martyana martyi</i> (Héribaud-Joseph) Round	13	0.3	0.2	2371.0	609.1
<i>Pseudostaurosira subsalina</i> (Hustedt) E.A.Morales	9	1.2	1.5	2368.9	691.5
<i>Cavinula scuttelloides</i> (W. Smith) Lange-Bertalot	4	0.0	0.1	2356.3	308.3
<i>Pseudostaurosira geocollegarum</i> (Witkowski) E.A.Morales	20	49.6	12.8	2347.8	505.0
<i>Navicula vulpina</i> Kützing	4	0.0	0.1	2313.7	259.6
<i>Aulacoseira granulata</i> (Ehrenberg) Simonsen	4	0.1	0.2	2311.1	310.3
<i>Karayevia clevei</i> (Grunow) Round & Bukhtiyarova	10	0.2	0.3	2310.3	656.3
<i>Fragilaria inflata</i> var. <i>istvanffy</i> (Pant.) Hustedt	4	0.0	0.1	2305.1	686.4
<i>Cocconeis placentula</i> var. <i>lineata</i> (Ehrenberg) Van Heurck	6	0.4	0.6	2290.1	576.6
<i>Entomoneis alata</i> (Ehrenberg) Ehrenberg	2	0.0	0.1	2282.4	102.0
<i>Amphora inariensis</i> Krammer	6	0.1	0.2	2281.2	395.3
<i>Belonastrum berlinensis</i> (Lemmermann) Round & Maidana	4	0.1	0.1	2273.6	245.3
<i>Stephanodiscus medius</i> Håkansson	3	0.3	0.7	2261.3	143.0
<i>Fragilaria sopotensis</i> Witkowski & Lange-Bertalot	3	0.5	1.1	2245.5	102.9
<i>Lindavia comensis</i> (Grunow) T.Nakov et al.	2	0.5	1.4	2243.3	517.1
<i>Placoneis placentula</i> (Ehrenberg) Mereschkowsky	3	0.0	0.1	2212.3	964.0
<i>Navicula rhynchocephala</i> Kützing	4	0.1	0.3	2211.7	805.6
<i>Cyclotella atomus</i> Hustedt	4	4.9	9.9	2186.6	174.1
<i>Rhoicosphenia abbreviata</i> (C.Agardh) Lange-Bertalot	4	0.1	0.1	2156.4	854.0
<i>Navicula radiosa</i> Kützing	3	0.0	0.1	2128.5	519.8
<i>Diatoma tenuis</i> C.Agardh	1	1.1	4.4	2091.3	560.3
<i>Stephanodiscus parvus</i> Stoermer & Håkansson	3	0.4	1.0	2077.8	364.1
<i>Ellerbeckia arenaria</i> (G.Moore ex Ralfs) R.M.Crawford	4	0.1	0.1	2059.5	514.4
<i>Planothidium lanceolatum</i> (Kützing) Round & Bukhtiyarova	2	0.0	0.1	2041.9	1014.5
<i>Navicula cryptocephala</i> Kützing	2	0.0	0.1	2038.6	869.5
<i>Fragilaria nanana</i> Lange-Bertalot	4	0.2	0.4	2006.3	349.2
<i>Actinocyclus normanii</i> (W.Gregory ex Greville) Hustedt	7	0.2	0.2	1944.8	658.2
<i>Hippodonta hungarica</i> (Grunow) Lange-Bertalot, Metzeltin & Witkowski	5	0.7	1.3	1921.9	643.2
<i>Navicula meniscus</i> Schumann	2	0.0	0.1	1906.9	613.3
<i>Gomphonema olivaceum</i> (Hornemann) Brébisson	2	0.0	0.1	1896.3	799.0
<i>Amphora libyca</i> Ehrenberg	4	0.3	0.7	1844.8	684.7
<i>Opephora olsenii</i> Møller	3	0.5	1.3	1780.2	769.0
<i>Navicula cincta</i> (Ehrenberg) Ralfs	3	0.0	0.1	1748.6	633.4
<i>Chaetoceros species</i>	2	0.0	0.1	1742.2	596.6
<i>Parlibellus crucicula</i> (W.Smith) Witkowski, Lange-Bertalot & Metzeltin	1	0.1	0.3	1556.1	799.0
<i>Planothidium delicatulum</i> (Kützing) Round & Bukhtiyarova	2	0.1	0.3	1470.7	799.0



importance to the functioning of these ecosystems. Environmental reconstructions based on diatoms may be useful for constructing salinity variability models, for example diatom-based salinity models from lakes in British Columbia (Cumming & Smol 1994) or the Thames Estuary (Juggins 1992). The salinity of Lake Resko Przymorskie is very diverse. The lake has the highest chloride levels compared to other Polish coastal lakes. Cieśliński (2011) showed that chloride concentration levels are on average above 1500 mg Cl<sup>-</sup> l<sup>-1</sup>, and the highest levels were determined in the western part of the lake, i.e. over 2200 mg Cl<sup>-</sup> l<sup>-1</sup>. Our study demonstrated that the chloride concentrations in the lake may be even higher. In November 2013, we measured concentrations ranging from 3090 mg Cl<sup>-</sup> l<sup>-1</sup> to 3231 mg Cl<sup>-</sup> l<sup>-1</sup>. Such high concentrations of Cl<sup>-</sup> unequivocally indicate that the lake is strongly saline.

According to the literature (Andrén 1999; Clarke et al. 2003; Weckström et al. 2002; Weckström et al. 2004; Kauppila et al. 2005; Weckström & Juggins 2005; and Weckström 2006), the distribution patterns of diatoms in the coastal area are also determined by nutrients concentration. The Gulf of Finland shows the effect of total nitrogen and total phosphorus on the structure of diatom assemblages dominated by small planktonic taxa (Weckström & Juggins 2005). In this study, *Rhoicosphenia curvata*, *Staurosirella pinnata*, *Opephora mutabilis*, *Navicula phyllepta*, *Bacillaria paxillifer* and *Pauliella taeniata* preferred lower nitrogen concentrations. Small centric planktonic taxa, such as *Cyclotella atomus*, *Cyclotella meneghiniana* and *Thalassiosira pseudonana*, were identified as indicators of highly elevated nutrient concentrations. These taxa could be used as potential indicators of the quality of coastal waters in the Gulf of Finland (Weckström & Juggins 2005; Weckström 2006). Hydrochemical and diatom analyses of Roskilde Fjord in Denmark and Laajalahti Bay in Finland demonstrated changes in nitrogen concentration and diatom abundance (mainly *Cyclotella choctawhatcheeana*) related to, among other factors, human activity (Kauppila et al. 2005; Clarke et al. 2006).

The nutrient status (especially NH<sub>4</sub><sup>+</sup> and PO<sub>4</sub><sup>3-</sup>) and heterogeneity of the habitat in this coastal lake is very specific. More than half of the studied sites have high values of these parameters. This is due to both the anthropogenic pollution of the lake and the impact and transformation of organic deposits into biogenic substances. Despite the high salinity of the lake, density stratification may not occur, because waters are completely mixed in shallow-water conditions. Findings of the research on coastal lakes (Søndergaard et al. 2001; Abesser & Robinson 2010; Cieśliński 2011; Woszczyk et al. 2014) indicate the possibility of mixing

of the surface bottom sediment layer. This is reflected in the high trophic status of the lake. One indication of this phenomenon observed during the study was the significant proportion of tychoplankton species in the pelagic zone of the lake.

### Ecological characteristic of diatoms in saline waters

We have shown that the species composition of diatoms is a sensitive indicator of salinity in Lake Resko Przymorskie, which suggests that diatom species data may be applied in paleoenvironmental studies. Findings related to a coastal lake have been presented for the first time, and our research involves the analysis of the diatoms' response to salinity gradients and the determination of chloride concentration preferences of selected diatom taxa.

The dominant diatom species was *Pseudostaurosira geocollegarum* (31.7-69.4% of the entire assemblage). It is typically a marine species (Morales 2002). According to the salinity classification by van Dam et al. (1984), *P. geocollegarum* can be included in a group of brackish diatoms, tolerating salinity levels of 1.8-9.0 PSU. The species occurs, however, in waters with salinity not higher than 7-8 PSU (Witkowski et al. 2000; Seddon et al. 2011), as confirmed by our results. Our research showed the optimum chloride level of more than 2300 mg Cl<sup>-</sup> l<sup>-1</sup>.

*Epithemia turgida* had the highest chloride optimum. According to the observations by Hofmann et al (2011), it is a common species in waters with a considerable concentration of electrolytes, and has been recorded not only in the brackish waters of Lake Resko Przymorskie but throughout the entire Baltic Sea (Hällfors 2004), with decreasing abundance or even absence toward the south (Snoeijis & Potapowa 1995), but identified in the Gulf of Gdańsk by Witkowski (1994), and in the brackish sediments of Kooser Wiesen, Körkwitz by Lampe & Janke (2002).

*Staurosira binodis* is a species distinguished by high vitality in a wide range of trophic states and electrolyte levels (Hoffmann et al. 2011). Moreover, *S. binodis* is also reported as a freshwater species that does not tolerate the full salinity gradient of the area from the Southern Baltic Proper, the Gulf of Gdańsk, the Gulf of Riga, and the Gulf of Finland (Hällfors 2004; Zgrundo et al. 2009). *Staurosira binodis* was also found in sediments of coastal lakes and their vicinity (Lutyńska 2008a; Mazurek et al. 2008; Witkowski et al. 2012; Sydor et al. 2015). Our studies have contributed to the knowledge about the ecology of this species.

Particularly high tolerance to chloride concentrations in the species structure of a given coastal lake

was found for such species as: *Amphora marina*, *Fragilaria capucina* var. *mesolepta*, *Placoneis placentula* and *Planothidium lanceolatum*. Besides its presence in Lake Resko Przymorskie, *A. marina* is widespread in the marine littoral zone (Witkowski et al. 2000). *Fragilaria capucina* var. *mesolepta* is a variety occurring in clean, slightly eutrophic waters (Krammer & Lange-Bertalot 1991a; Arrhenius et al. 2014) and was reported from the Gulf of Riga (Hällfors 2004). *Planothidium lanceolatum* is a species reported from the Southern Baltic, the Gulf of Riga, the Gulf of Finland (Hällfors 2004) and the Gulf of Gdańsk (Witkowski 1994).

Indicator species of the lowest salinity (below 1600 mg Cl<sup>-1</sup>) within the range of Lake Resko Przymorskie included *Parlibellus crucicula* and *Planothidium delicatulum*. *Parlibellus crucicula* occurs on marine coasts, in river estuaries, and in saline coastal marshes (Krammer & Lange-Bertalot 1997). It mostly occurs in the littoral zone and is reported from the Gulf of Riga and the Gulf of Finland (Hällfors 2004) and the Gulf of Gdańsk (Witkowski 1994; Zgrundo et al. 2009). *Planothidium delicatulum* is a species occurring in brackish inland waters and on the seashores (Witkowski et al. 2004; Krammer & Lange-Bertalot 1991b). It is reported from the Southern Baltic, the Gulf of Riga, the Gulf of Finland and the Gulf of Bothnia (Hällfors 2004). Moreover, it is abundant in the littoral and sub-littoral zone of the Gulf of Gdańsk (Witkowski 1994).

Turbid nutrient-rich areas of Lake Resko Przymorskie were dominated by tychoplanktonic species, classified as tychoplanktonic of eponitic origin and benthic origin. In July 2013, an increased contribution of planktonic species was observed, including mainly *Cyclotella meneghiniana* and *Cyclotella atomus*, which were also observed in high-nutrient waters of Chesapeake Bay (Cooper 1995), Florida Bay (Wachnicka et al. 2010) and the Baltic Sea (Andrén et al. 2000; Weckström & Juggins 2005; Zgrundo et al. 2009). Our research showed an increase in the abundance of *Cyclotella atomus* in July 2013 samples, which correlates with temperature, whereas *Cyclotella meneghiniana* occurs in samples with higher concentrations of phosphorous. Small planktonic taxa such as *Cyclotella atomus* and *Cyclotella meneghiniana* may be useful indicators of highly elevated concentrations of nutrients (Weckström & Juggins 2005).

## Conclusions

Our research in Lake Resko Przymorskie revealed that: (1) the parameters controlling the species structure of diatoms include chloride levels, water

temperature, pH and phosphate content, (2) the composition and structure of diatom assemblages along the spatial and temporal salinity gradient are related to the inflow of marine waters, and (3) the dominance of species preferring higher salinity, particularly *Pseudostaurosira geocollegarum*. The system of Lake Resko Przymorskie is strongly dependent on marine water intrusion via the northern outlets and freshwater inflow from the drains and canals that connect with the lake.

The structure of diatom species composition during the study period showed a predominance of brackish species. Such a diatom species structure suggests a constant inflow of saline waters into Lake Resko Przymorskie. Increased salinity caused the development of a diatom species composition specific to the lake, with individual optima and a very narrow tolerance to chloride content (e.g., *Epithemia turgida* and *Staurosira binodis*). Our findings on greater chloride tolerance of taxa living in the waters of Lake Resko Przymorskie are not final and will require further research involving other coastal lakes.

The results of our research will provide the basis for the development of training sets for coastal lakes useful in environmental reconstructions using the transfer function.

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