

Diversity of the Rhopalodiaceae Diatoms (Bacillariophyta) on Macrophytes of Different Architecture in Small and Shallow Oxbow Lakes (SE Poland)

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ABSTRACT

The research was conducted in July 2018 on six oxbow lakes of the San River. A total of 21 samples from various macrophyte species were collected. Over 400 species of diatoms were identified in the analyzed materials, 9 of which belonged to the Rhopalodiaceae family. Most of these *Epithemia* and *Rhopalodia* species developed as dominants in most of the studied samples. One of reported species – *Epithemia selengaensis* Vishnyakov, Kulikovskiy & Genkal was first time reported from Poland. The aim of the study was to present the taxonomic diversity of the representatives of the Rhopalodiaceae family growing different macrophyte species in oxbow lakes in the summer aspect.

Keywords: epiphytic algae, *Epithemia*, *Rhopalodia*, macrophytes, ecology, San River, taxonomy

INTRODUCTION

Communities of primary producers (macrophytes, phytoplankton and benthic algae) play a crucial role in supporting various food webs within the aquatic ecosystem (Špoljar et al. 2017). Nevertheless, the benthic algal communities (including epiphytic) represent an important component of food webs as primary producers, particularly in oligo-mesotrophic shallow lakes or littoral zones, where high water transparency results in higher light availability for microorganisms living below the water surface (Sierszen et al. 2006). Apart from its ecological role, algal epiphytic communities are also used in the assessment of the ecological status of aquatic ecosystems. It applies primarily to diatoms, which fulfill many the requirements of being useful indicator organisms in running waters, although several studies

showed that the diatom analyses may also be applied to determine the ecological status of lentic ecosystems, especially shallow lakes (Kitner, Poulícková 2003, Cejudo-Figueiras et al. 2011, Blanco et al. 2014, MacDougall et al. 2017). It seems to be broadly accepted that the bioassessment methods based on epiphytic diatoms may be also effective for tracking the past changes under the environmental conditions of lakes or even small ponds (Wiklund et al. 2010, Emson et al. 2018).

The studies concerning epiphytic diatoms in small and shallow water bodies like ponds or oxbow lakes are relatively scarce, despite the fact that small waterbodies play an important role in the structure of biodiversity on both landscape and species level (Scheffer et al. 2006, Cereghino et al. 2008). The importance of oxbow lakes in Europe as biodiversity hot-spots was acknowledged by EC Habitats Directive

92/43/EEC; thus, they became protected under the name “Habitat 3150” (Nowak et al. 2015). Their presence in the river valleys enriches biodiversity as habitats form protected, rare, valuable and disappearing species of vascular plants (Michalska-Hejduk et al. 2009) but also other organisms. In oxbow lakes, which usually are small and shallow water bodies, macrophytes play a crucial role in the ecosystem functioning; thus, the structure of aquatic plants is the main indicator which is used in the bioassessment of their conservation status (Wilk-Woźniak et al. 2019). As other shallow lake ecosystems with differentiated macrophyte architecture they offer an excellent habitat for epiphytic diatom assemblages (Letáková et al. 2018).

Rhopalodiaceae (Karsten) Topachevs’kyj and Oksiyuk (Round et al. 1990) are one of only two families within the Rhopalodiales order. Among several small genera only two: *Rhopalodia* and *Epithemia* contain several dozen of species. The most characteristic feature of species within this two genera is the presence of 2–5 cyanobacterial endosymbionths (Geitler 1977, Janson 2002). The cell walls of both genera are similar in symmetry, being strongly dorsiventral. The raphe of *Rhopalodia* species is positioned on the dorsal side, in a shallow keel, whereas the raphe of the species within the *Epithemia* genus is positioned along the ventral margin, with simple or slightly expanded raphe endings. In contrast to *Epithemia*, *Rhopalodia* cells have no girdle bands (Round et al. 1990, Taylor, Cocquyt 2016). Most of the *Epithemia* species are cosmopolitan and have worldwide distribution, although generally prefer temperate climates. They are widespread in almost all freshwaters, growing on submerged vegetation or more seldom in epilithic assemblages (Sims 1983, Krammer, Lange-Bertalot 1988). The *Epithemia* species reaches the maximum abundance especially in those microhabitats where phosphorus is relatively more available (DeYoe 1992, Kociolek et al. 2015).

The representatives of the Rhopalodiaceae family, due to the large cell size and wide geographical distribution, are well known species, often described more than 100 years ago. However, the taxa belonging to the two discussed genera, are usually identified only to the species level, based on a broad species concept. As a result the real diversity and within the family is often underestimated.

The paper discusses the variability and occurrence of the diatom taxa from the *Rhopalodiaceae* family from six oxbow lakes in the San river catchment area. All noted species developed on the submerged macrophytes. Additional information about morphology, ecology of *Rhopalodia* and *Epithemia* taxa based on observed populations was provided.

STUDY AREA

The San River, the largest of Carpathian rivers, is 458 km long. It is also one of the main tributaries of Vistula River. Oxbow lakes are very numerous in the San catchment area, especially along the middle and lower sections of the river.

The study was conducted in six oxbow lakes formed by the San River (south-eastern Poland) in its middle (sites P1, P2, M1, H1) and lower (Z1, Z2) course (Fig. 1). First two (P1 and P2) are located in the outskirts of the Przemyśl city (49°47’N, 22°46’E). These water bodies with an area of 0.22 and 0.11 ha, are part of a larger group, located close to the San River, in the vicinity of allotment gardens. The area of both water bodies is overgrown with aquatic and water-side vegetation. Another two study sites (H1 and M1) are situated east of Przemyśl, close to the state border with Ukraine. The largest of studied oxbow lakes in the village Hurko (H1), with an area of ca. 10 ha, is used for fishery. The site M1 is a part of vast park surrounding the old historical palace in Medyka village, thus playing a role of a landscape pond. Both lakes (H1, M1), being under the influence of the human activity, have rather scarce aquatic vegetation, which was removed during its management. During the field studies in these lakes, a cyanobacterial bloom was observed. The sites Z1 and Z2, situated in the lower course of the San River have the most natural character. They are located within the NATURA 2000 Special Area of Conservation “Dolina Dolnego Sanu” (PLH180020), situated between Stalowa Wola (50°34’N, 22°03’E) and Sandomierz (50°40’N, 21°44’E), close to the village Zaleszany. Both sites are part of so-called “Old San”, the largest of the river-beds in the lower course of the San River, with several arms stretched between two present river beds (San and Łęg rivers) (Prarat 2009, Michalska-Hejduk et al. 2009).

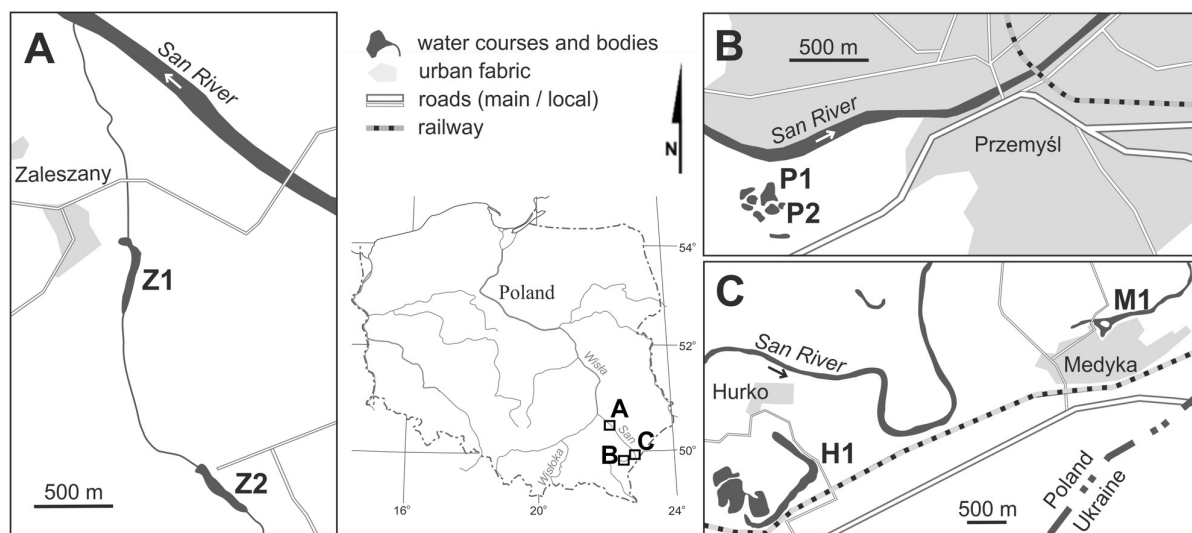


Figure 1. Situation map of six studied oxbow lakes

METHODS

The diatom material was collected in July 2018 from six different oxbow lakes in four localities. The samples were taken from dominant macrophytes (floating leaves (FLM), emergent (EM) and submerged plants (SM)). A total of 21 samples were collected, separately for each of the macrophyte species (Table 1). The plant species identification was based on Kłosowski, Kłosowski (2010).

Water temperature, oxygen concentration, pH and conductivity were measured in situ during sampling, with the use of a MARTINI PH65 meter, a MARTINI EC59 meter and a HANNA HI 9146 oxygen meter, respectively. An additional water sample for the chemical analyses (listed in Table 2) was taken from the central part of each oxbow lake. Those analyses were performed using a DIONEX ICS-5000 + DC Thermo scientific ion chromatograph in a laboratory.

The diatom samples were cleaned by adding 30% H_2O_2 and heating until the organic matter is digested (1–2 h). The reaction was completed by the addition of HCl. Following digestion and centrifugation (5 times, 5 minutes at 2500 rpm) with distilled water, the resulting cleaned material was mounted in Pleurax (refractive index 1.75). The diatoms were identified and counted under a Carl Zeiss Axio Imager A2 light microscope (LM) equipped with Differential Interference Contrast (Nomarski) optics, under 1000 \times magnification. The diatom images were captured using a Zeiss ICC 5 camera. The diatom identification was based on: Hofmann et al. (2011), Lange-Bertalot

et al. (2017), Kulikovskiy et al. (2016) and Qingmin et al. (2009). The species composition was determined by counting 300 valves on randomly selected transects under LM.

Recently, Cocquyt et al. (2018) degraded the *Rhopalodia* genus to the rank of subgenus in the *Epithemia* genus, but for the purposes of this study, the authors used the classification of genera provided by Round et al. (1990).

RESULTS

The values of physical and chemical parameters measured in the study are shown in Table 2. Temperature (23.8–26.3 °C) and pH values (7.4–8.5) fell within the ranges expected for the oxbow lakes in Central Europe in summer. Some differentiation among the studied lakes were observed in electrolytic conductivity. The lowest values of conductivity were measured at sites Z1 and Z2 in Zaleszany (347–367 $\mu S cm^{-1}$). The rest of lakes had conductivity in the range 500–565 $\mu S cm^{-1}$ except P2 site in Przemyśl, where twice as high values of this parameter were found, as compared to Z1 and Z2 sites (Table 2). The water in all studied lakes was rich in chlorides (17.72–67.54 $mg l^{-1}$) and calcium (26.5–83.3 $mg l^{-1}$) but had very low values (below the limit of determination) of nitrates and phosphates (with the exception of site M1).

More than 400 diatom taxa were identified in 21 samples. The number of taxa noted in each sample ranged from 40 (sample 2018/152 collected from *Potamogeton lucens* at site P1) to 137

Table 1. List of samples used in this study with detailed information about sites, geographic coordinates and the macrophytes samples were taken from.

Site	Samples	GPS data	Vegetation	
Medyka (M1)	2018/144	49°48'31.9"N 22°55'58.6"E	<i>Ceratophyllum demersum</i> L.	SM
	2018/146		<i>Phragmites australis</i> (Cav.) Trin. ex Steud.	EM
	2018/147		<i>Lemno-Spirodeletum</i> (Koch, 1954)	FLM
Hurko (H1)	2018/142	50°39'24.6"N 21°55'21.7"E	<i>Phragmites australis</i> (Cav.) Trin. ex Steud.	EM
Przemysł (P1)	2018/150	49°46'39.8"N 22°44'41.0"E	<i>Hydrocharis morsus-ranae</i> L.	FLM
	2018/151		<i>Typha</i> sp.	EM
	2018/152		<i>Potamogeton lucens</i> L.	SM
Przemysł (P2)	2018/154	49°46'38.0"N 22°44'44.6"E	<i>Hydrocharis morsus-ranae</i> L.	FLM
	2018/155		<i>Potamogeton lucens</i> L.	SM
	2018/156		<i>Myriophyllum spicatum</i> L.	SM
	2018/157		<i>Typha</i> sp.	EM
	2018/158		<i>Utricularia australis</i> R. Br.	SM
Zaleszany (Z1)	2018/131	50°40'07.1"N 21°54'54.9"E	<i>Hydrocharis morsus-ranae</i> L.	FLM
	2018/133		<i>Salvinia natans</i> (L.) All.	FLM
	2018/134		<i>Nuphar lutea</i> (L.) Sibth. & Sm.	FLM
	2018/135		<i>Myriophyllum spicatum</i> L.	SM
	2018/136		<i>Ceratophyllum demersum</i> L.	SM
Zaleszany (Z2)	2018/137	50°39'24.6"N 21°55'21.7"E	<i>Hydrocharis morsus-ranae</i> L.	FLM
	2018/139		<i>Salvinia natans</i> (L.) All.	FLM
	2018/140		<i>Nuphar lutea</i> (L.) Sibth. & Sm.	FLM
	2018/141		<i>Phragmites australis</i> (Cav.) Trin. ex Steud.	EM

(sample 2018/146) on *Phragmites australis* in Medyka (site M1) (Table 3). Among all recorded taxa, nine belong to the Rhopalodiaceae family, from which seven *Epithemia* and two *Rhopalodia* taxa were noted in all studied samples, but in variable abundance (Table 3). At the sites devoid of their „semi-natural” character (H1, M1), the Rhopalodiaceae taxa were noted only occasionally, whereas in macrophyte dominated oxbow lakes of more “natural” character (P1, P2, Z1, Z2), they developed more numerously. The diatoms from that family dominated the assemblages in some cases: in six from twenty one studied

samples, Rhopalodiaceae reached together more than 50% off all counted valves. Among them, in the sample collected from *Myriophyllum spicatum* (2018/135), the diatom community was entirely dominated by *Epithemia* and *Rhopalodia* taxa. However, no preferences of individual taxa in relation to macrophytes were observed.

Below, the population characteristics of Rhopalodiaceae taxa found in the study are presented.

Epithemia adnata* var. *adnata (Kützing) Rabenhorst (Fig. 2: 7–10)

Population characteristic: length 23.2–87.4 μm, width 7.4–10.3 μm, striae 14–16 in 10 μm, costae

Table 2. Physico-chemical parameters of water in the six studied oxbow lakes.

Sites	M1	H1	P1	P2	Z1	Z2
Temperature [°C]	25.3	26.3	23.8	25.4	26.1	24.5
pH	8.5	7.9	7.7	7.4	7.7	7.4
Conductivity [μS cm ⁻¹]	523	500	565	728	347	367
O ₂ [mg l ⁻¹]	5.68	4.86	6.10	2.15	1.54	1.87
Cl ⁻ [mg l ⁻¹]	67.54	25.11	28.40	25.58	20.38	17.72
SO ₄ ²⁻ [mg l ⁻¹]	15.94	26.25	19.19	13.44	3.07	14.22
PO ₄ ³⁻ [mg l ⁻¹]	0.47	<0.001	<0.001	<0.001	<0.001	<0.001
NO ₃ ⁻ [mg l ⁻¹]	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001
NH ₄ ⁺ [mg l ⁻¹]	0.46	0.21	0.18	0.15	0.24	0.45
Mg ²⁺ [mg l ⁻¹]	8.03	15.05	12.26	12.48	7.47	6.63
Ca ²⁺ [mg l ⁻¹]	26.52	44.52	56.22	83.26	30.49	28.61

Table 3. The presence and percentage of counted valves in diatom assemblages of studied taxa. The highest values are in bold, + taxon observed occasionally, reaching up to 1%, T – total number of identified taxa, EAVA – *Epithemia adnata* var. *adnata*, EAVP – *Epithemia adnata* var. *proboscidea*, EAVS – *Epithemia adnata* var. *saxonica*, EFRI – *Epithemia frickei*, ESEL – *Epithemia selengaensis*, ESOR – *Epithemia sorex*, ETUR – *Epithemia turgida*, RGVG – *Rhopalodia gibba* var. *gibba*, RGVM – *Rhopalodia gibba* var. *minuta*.

Samples		T	EAVA	EAVP	EAVS	EFRI	ESEL	ESOR	ETUR	RGVG	RGVM
M1	<i>Ceratophyllum demersum</i>	108	+						+		
	<i>Phragmites australis</i>	137	+	+				+	+	+	
	<i>Lemno-Spirodeletum</i>	75	+	+			+		+	+	
H1	<i>Phragmites australis</i>	73	+	+		+	+	+	+	+	
P1	<i>Hydrocharis morsus-ranae</i>	88	33.6	8.9				+	2.5	9.5	
	<i>Typha</i> sp.	123	2.2	28.4	+			2.2	1.5	12.7	
	<i>Potamogeton lucens</i>	40	36.6	13.4				12.5	3.5	22.3	
P2	<i>Hydrocharis morsus-ranae</i>	72	14.1	10.9					+	1.6	
	<i>Potamogeton lucens</i>	42	13.2	28.5				+	4.9	31.9	+
	<i>Myriophyllum spicatum</i>	81	15	12.9					+	4.1	
	<i>Typha</i> sp.	84	8	4.5	+		+		+	2.3	
	<i>Utricularia australis</i>	69	+	16.8					+	17.5	
Z1	<i>Hydrocharis morsus-ranae</i>	65	11.7	+		+		+	5.6	+	
	<i>Salvinia natans</i>	76	8.4	+		+		+	3.8	8.4	
	<i>Nuphar lutea</i>	68	31.3	+	+			+	+	20.9	
	<i>Myriophyllum spicatum</i>	50	21.9	+		+		+	10.5	66.6	+
	<i>Ceratophyllum demersum</i>	62	28.8	+		+			5.1	5.1	
Z2	<i>Hydrocharis morsus-ranae</i>	89	10.3	4.2				+	+	4.1	
	<i>Salvinia natans</i>	93	+	+	+			+	+	+	
	<i>Nuphar lutea</i>	56	23.8	23.8				+	24.6	6.9	
	<i>Phragmites australis</i>	50	13.4	20.3	+	+		+	5.9	2.7	+

3–8 in 10 µm. The taxon was observed in all studied samples, often formed numerous population. In the reservoirs that keep their natural character (P1, P2, Z1, Z2) it dominates the diatom flora, and reached up to 36.6% of total diatom abundance in the sample 2018/152. On the contrary, at sites M1 and H1, this variety was observed only occasionally.

Epithemia adnata var. *proboscidea* (Kützing) Hende (Fig. 2: 1–5)

Population characteristic: length 26.7–100 µm, width 7.4–10.2 µm, striae 14–16 in 10 µm, costae 3–8 in 10 µm. The taxon co-occurs with nominal form in all studied samples, often formed numerous populations (up to 28.5% of total diatom abundance in sample 2018/155). Only at sites M1, H1, Z1 in Zaleszany, it was less frequent.

Epithemia adnata var. *saxonica* (Kützing) R.M.Patrick (Fig. 3: 13–16)

Population characteristic: length 25.5–90.2 µm, width 8–9.4 µm, striae 12–15 in 10 µm, costae 3–6 in 10 µm. This variety was observed only in form of individual specimens together with other varieties of *E. adnata*.

Epithemia frickei Krammer in Lange-Bertalot & Krammer (Fig. 2: 6)

Population characteristic: length 27–85 µm, width 10.4–12.4 µm, striae 12–13 in 10 µm, costae 2–4 in 10 µm. Only few specimens were observed in the samples from Zaleszany (both sites) and Hurko. Observed valves were large, reached the upper range of dimensions for this taxa.

Epithemia selengaensis Vishnyakov, Kulikovskiy & Genkal (Fig. 2: 11–12)

Population characteristic: length 30–58 µm, width 8–12 µm, striae 10–12 in 10 µm, costae 2–3 in 10 µm. Single specimens of these species were observed in only three samples from upper part of San river, both in lakes subjected to the anthropogenic pressure (H1, M1) and the natural one (P2).

Epithemia sorex Kützing (Fig. 3: 8–12)

Population characteristic: length 10–45 µm, width 7–8.2 µm, striae 13–16 in 10 µm, costae 5–7 in 10 µm. The species was found in each of studied oxbow lakes, but mainly occasionally. Only at site P1 it formed larger population on *Potamogeton lucens* (12.5% of all counted valves).

Epithemia turgida (Ehrenberg) Kützing (Fig. 3: 1–3)

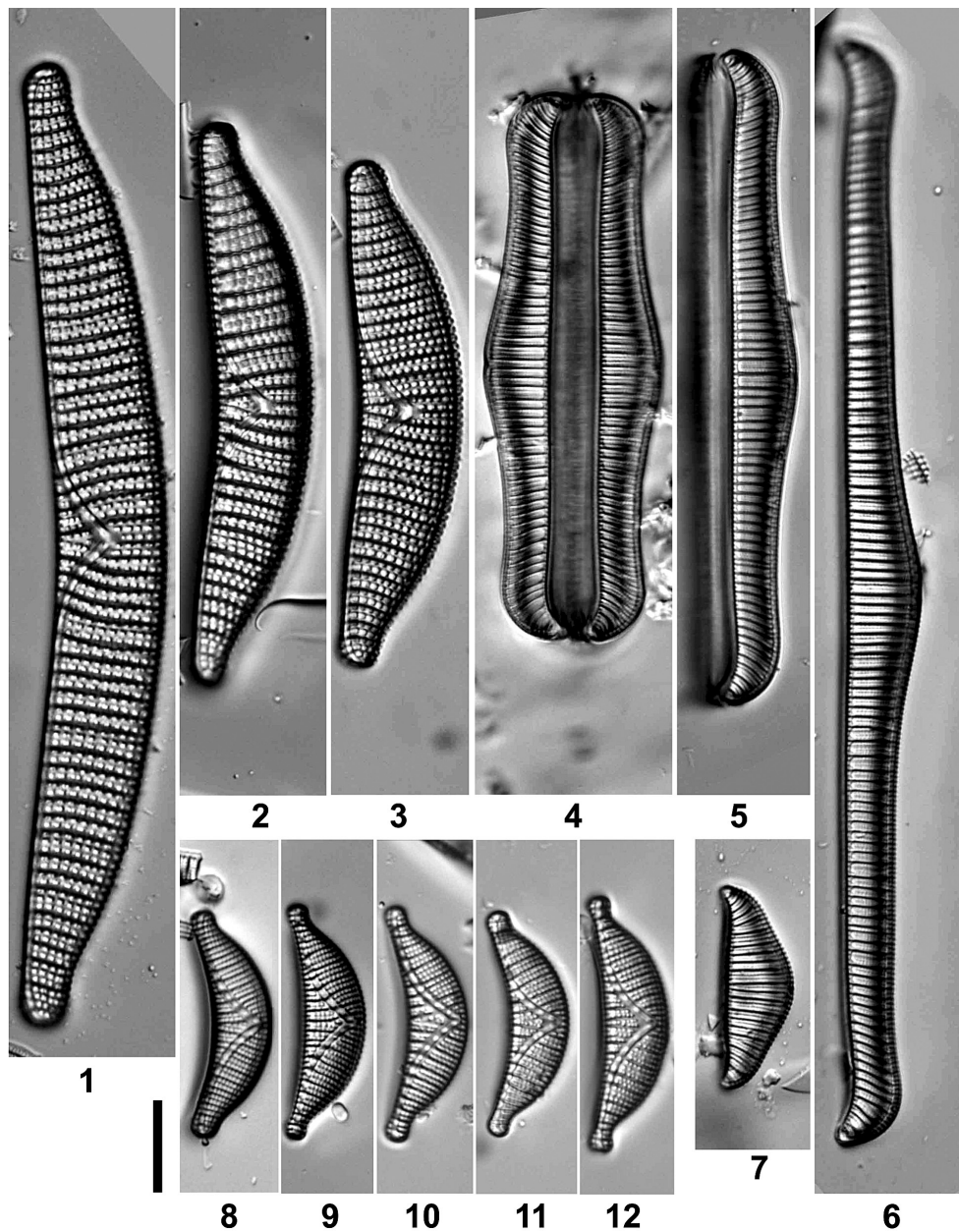


Figure 2. LM pictures of *Epithemia* and *Rhopalodia* taxa recorded in samples collected from different macrophytes. 1–3 – *Epithemia turgida*, 4–6 – *Rhopalodia gibba*, 7 – *Rhopalodia gibba* var. *minuta*, 8 – *Epithemia soresx*.

Population characteristic: length 44–140.4 μm , width 14–17.8 μm , striae 7–8 in 10 μm , costae 3–5 in 10 μm . The species was observed in all studied samples, usually reached few percent of all valves counted in each sample. Only in sample 2018/140, the species was more numerous (24.6% of all counted valves). This sample was collected from *Nuphar lutea*.

Rhopalodia gibba (Ehrenberg) O.Müller (Fig. 3: 4–6)

Population characteristic: length 10–210 μm , width 8.7–11 μm , striae 14–16 in 10 μm , costae

6–8 in 10 μm . The species was observed in almost all studied samples (with exception of sample 2018/144), where often formed numerous populations (exceeding 5% share in an assemblage). The most frequent population was noted in the sample collected from *Myriophyllum spicatum*.

Rhopalodia gibba* var. *minuta Krammer (Fig. 3: 7)

Population characteristic: length 25–29 μm , width 8.4–8.9 μm , striae 14–16 in 10 μm , costae 7–8 in 10 μm . Only few specimens were observed together with nominal variety.

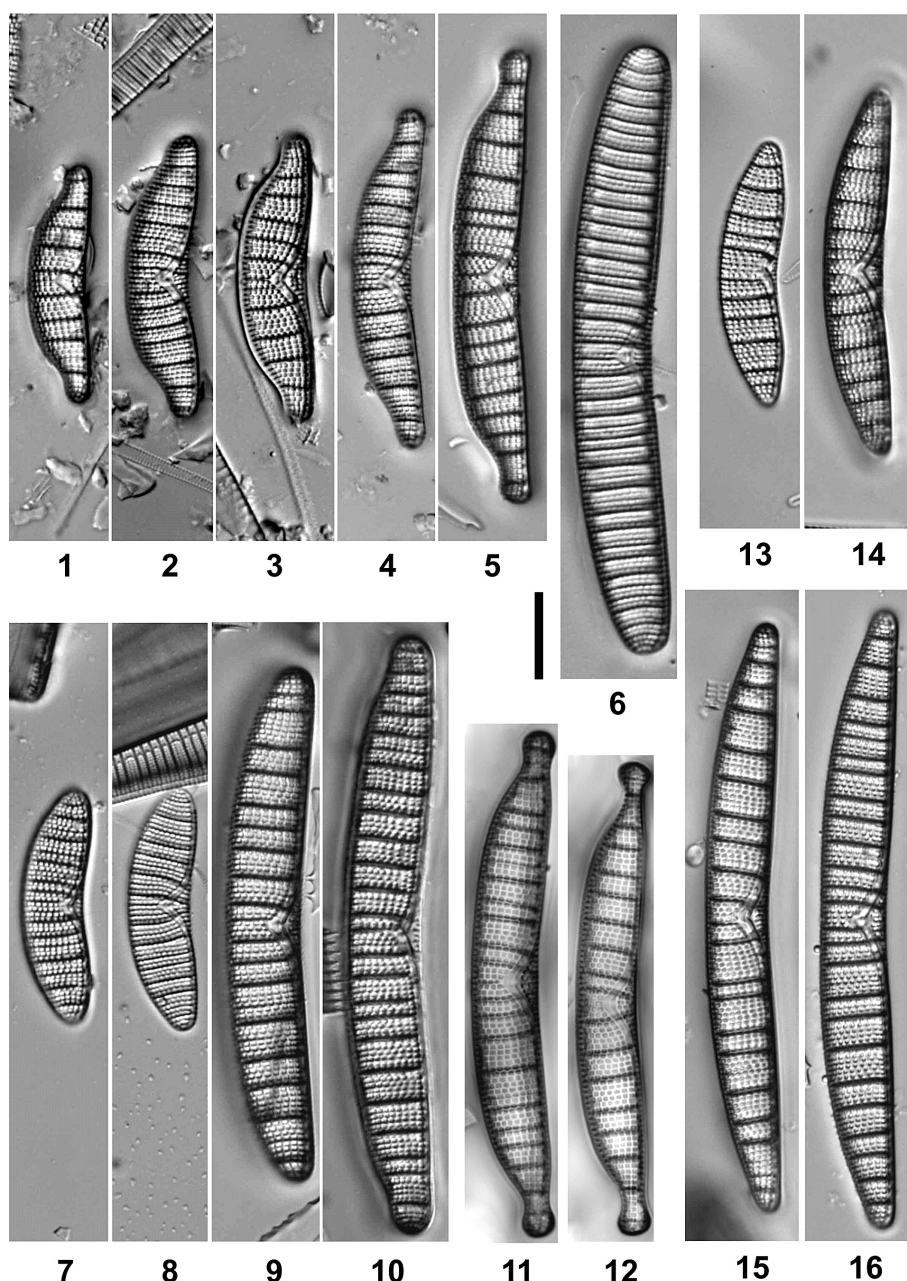


Figure 3. LM pictures of *Epithemia* taxa recorded in samples collected from different macrophytes. 1–5 – *Epithemia adnata* var. *proboscidea*, 6 – *Epithemia frickei*, 7–10 – *Epithemia adnata* var. *adnata*, 11–12 – *Epithemia selengaensis*, 13–16 – *Epithemia adnata* var. *saxonica*

DISCUSSION

The oxbow lakes can develop as a result of isolation/disconnection from meander necks or separation from rivers by riverside embankments during freshets (Żmudziński et al. 2002, Dembowska, Napiórkowski 2012, Dembowska et al. 2012). The oxbow lake has many important functions in river valley ecosystems, where together with rivers and their catchments it contributes to the natural diversity of those areas. Apart their hydrological and microclimatic role of retention

reservoirs, oxbow lakes form a suitable and supportive environment for the growth of water and hygrophilous flora, as well as valuable habitat for wildlife (Glińska-Lewczuk 2009, Koc et al. 2009).

In general, in all studied oxbow lakes more or less similar values of water temperature, pH and conductivity were noted, which is in accordance with the results presented by Michalska-Hejduk et al. (2009) in the same locality. Only the oxygen content was higher (2.52–4.21 mg/l) than in our samples, which is probably connected with differences in the weather conditions and water temperature.

Oxbow lakes are considered as natural eutrophic water bodies. However, as a result of macrophyte and phytoplankton activities, the levels of soluble mineral forms of nutrients in such lakes can be very low, as it was noted in the studied lakes (Reddy et al. 1989, 1990, Olguín et al. 2007, Ozimek et al. 1990, Upadhyaya et al. 2007). The low values of the analyzed ions might also be a result of the oxbow separation from the river, as it was revealed by Glińska-Lewczuk (2009). In her study, in the comparison of two types of oxbow lakes: lentic and lotic, it was demonstrated that various ions are withdrawn from the system. The oxbow lakes separated from the river showed significantly lower values of nitrates and nitrites. In lentic oxbow lakes, isolated from the river, the matter cycling (mainly organic) influences the change and development of the specific macrophytes species composition. The plant species in this type of habitat are well adapted to the anoxic conditions and decreased productivity (Glińska-Lewczuk 2009).

From seven identified in this study *Epithemia* species, *E. selengaensis* was reported for the first time from Poland. The species was described in 2014 from Selenga River delta in Russia, but was also observed in the samples from lakes in Mongolia and Eastern Baikal Region (Vishnyakov et al. 2014). The species was also recorded on macrophytes in various types of water in Turkey (C. Solak – pers. comm.). *E. selengaensis* belongs to *E. adnata* group but can be easily distinguished on the basis of roundly capitate apices. Other Rhopalodiaceae noted during our study belong to commonly reported taxa. Unfortunately, all of them are identified in wide species concept, very often only to the species level. They are widespread in fresh and brackish waters and prefer waters rich in calcium, meso- to eutrophic. They occur mainly in epiphytic, more seldom in epilithic assemblages in stagnant and slowly floating waters (Hofmann et al. 2011, Lange-Bertalot et al. 2017, Pliński, Witkowski 2013).

The studied taxa were found on each of the analyzed macrophytes. The most numerous population was formed by *Epithemia adnata* s.l., followed by *E. turgida* and *Rhopalodia gibba* s.l. The semi-natural oxbow lakes (i.e. in Przemyśl city) overgrown by various macrophytes favor the development of epiphytic diatom species. The stagnant, freshwater ecosystems in Europe are very often overgrown by macrophytes. Among the epiphytic diatom assemblages growing on various macrophytic plants, many authors report

the dominance of species from the Rhopalodiaceae, especially *E. adnata* s.l., *E. sorex*, *R. gibba* (Kuczyńska-Kippen et al. 2005, Laugaste, Reunanen 2005, Toporowska et al. 2008). In the samples taken from the lakes where the cyanobacterial bloom persisted (site H1, M1), the discussed species developed rarely and assemblages were mainly dominated by typically epiphytic taxa. On stems of *Phragmites australis* and on *Ceratophyllum demersum* the most common taxa were: *Cocconeis placentula* Ehrenberg, *C. lineata* Ehrenberg and *Cymbella subleptoceros* Krammer. In plant community “*Lemno-Spirodeletum*” diatom assemblages was dominated by *C. lineata* Ehrenberg, *Halamphora veneta* (Kützing) Levkov and *Lemnicola hungarica* (Grunow) Round & Basson. Moreover, *Lemnicola hungarica* is reported as a species associated with various Lemnaceae species (Buczko 2007, Emson et al. 2018).

In addition to the dominance of macrophytes, also the nitrogen limitation tends to give preferential conditions for the Rhopalodiaceae diatoms (Fairchild et al. 1985, Hill, Knight 1988, Peterson, Grim 1992). This species have endosymbionts capable of fixing the atmospheric nitrogen. Marks, Lowe (1993) reported that *E. adnata* is a nitrogen specialist, often dominates epiphytic microhabitats where the ratio of available nitrogen to phosphorous might be relatively low.

The *Epithemia* and *Rhopalodia* genera contain mainly the species considered as polytypic, containing many varieties. However, in most commonly used keys (Hofmann et al. 2011, Lange-Bertalot et al. 2017), only the broad species concept is given for most of them, resulting in poor understanding of ecological preferences and the distribution of individual taxa of the *Epithemia* and *Rhopalodia* genera. Therefore, the actual diversity of the Rhopalodiaceae family is also overlooked.

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