Wpływ wybranych czynników środowiskowych na różnorodność okrzemek (Bacillariophyta) oraz na wartości wskaźników okrzemkowych jezior Wigierskiego Parku Narodowego

Effect of selected environmental factors on diatoms' diversity (Bacillariophyta) and on the values of diatom indexes of the lakes of the Wigry National Park

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2. **Eliasz-Kowalska, M.**, Wojtal, A. Z., Barinova, S. Influence of Selected Environmental Factors on Diatom β Diversity (Bacillariophyta) and the Value of Diatom Indices and Sampling Issues. *Water* **2022**, *14* (15), 2315. https://doi.org/10.3390/w14152315. IF=3.530, MEiN=100.

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STRESZCZENIE

Okrzemki (Bacillariophyta) są uważane za jedne z najlepszych indykatorów stosowanych do oceny statusu ekologicznego wód powierzchniowych. Ramowa Dyrektywa Wodna wymaga od użytkowników klasyfikowania wszystkich wód powierzchniowych według wskaźnikowego statusu ekologicznego, co daje bardziej holistyczne spojrzenie na ocenę stanu danego ekosystemu. Zbadany stan ekologiczny wody powierzchniowej pozwala na odpowiednie działania zapobiegawcze przed pogorszeniem warunków środowiskowych. Jednak wiele krajów, w tym Polska, nadal ma problem z precyzyjną interpretacją wskaźników okrzemkowych w jeziorach. Wiele krajów jest również w trakcie wprowadzania wskaźników ekologicznych (np. okrzemkowych) do oceny ekosystemów wodnych. Wpływ antropopresji i zmian klimatu doprowadziły do postępującego spadku różnorodności okrzemek w jeziorach. Składniki różnorodności β (np. wymiana gatunków lub ich zagnieżdżenie) i czynniki wpływające na zmiany różnorodności β są nadal słabo poznane, mimo że różnorodność odgrywa ważną rolę w wyjaśnianiu procesów ekologicznych. Zrozumienie mechanizmów odpowiedzialnych za zmiany w składnikach różnorodności β (zagnieżdżenia i wymiany gatunków) wraz z gradientami czynników środowiskowych jest obecnie głównym problemem w biologii i ochronie wielu gatunków. Oszacowanie β różnorodności pozwala na zmierzenie różnic między badanymi zbiorowiskami. Przyczynia się także do zrozumienia różnych poziomów oddziaływania warunków środowiskowych i jakości wody na zbiorowiska wodne.

Celem rozprawy doktorskiej było zbadanie wpływu wybranych czynników środowiskowych na różnorodność gatunkową okrzemek oraz na wartości wskaźników okrzemkowych jezior Wigierskiego Parku Narodowego.

W pracy sprawdzono, w jaki sposób wybrane czynniki środowiskowe wpływają na zbiorowiska okrzemkowe. Analizowano **(w art. 1: Diversity 2020)** w jaki sposób różnice środowiskowe między jeziorami a) dysharmonijnymi, b) harmonijnymi z większą antropopresją i c) harmonijnymi z ograniczonym wpływem człowieka wpływają na zbiorowiska okrzemkowe. Znaczące różnice były widoczne w indeksie α Wskaźniku Dominacji. Wykazano, że jeziora dysharmonijne, harmonijne z większą antropopresją oraz

z mniejszą antropopresją charakteryzują się różnymi czynnikami środowiskowymi, szczególnie pH, stężeniem jonów chlorkowych i jonów siarczanowych co jest związane z różną strukturą dominacji. Zbadano **(w art. 2: Water 2022)** różnice w różnorodności β i Indeksu Okrzemkowego Jezior (IOJ) oraz reakcje okrzemek na wybrane czynniki środowiskowe. Zbadano również reprezentatywność próbek w szacowaniu stanu ekologicznego jeziora. Wyniki wyraźnie wskazują, że zwiększenie dokładności oceny środowiska może zostać osiągnięte poprzez dokładnie oznaczanie okrzemek do gatunków w stosunku do podejścia grupowania gatunków. Dodatkowo w artykule 3 precyzyjne badania taksonomiczne wskazują i dokumentują, jak cenne i bogate w gatunki rzadkie i chronione są jeziora Wigierskiego Parku Narodowego **(w art. 3: maszynopis)**.

Praca dotyczy analizy zmian zbiorowisk okrzemkowych w gradientach środowiskowych ze szczególnym uwzględnieniem różnorodności α i β oraz ze zmianami wartości Indeksu Okrzemkowego Jezior. Wyniki badań dotyczą różnic w różnorodności α i β w zależności od antropogenicznych zmian w ekosystemach wodnych. Zmiany środowiskowe powinny być odpowiednio mierzone szczególnie w najbardziej wrażliwych ekosystemach słodkowodnych.

SUMMARY

Diatoms (Bacillariophyta) are considered to be one of the best indicators used to assess the ecological status of surface waters. The Water Framework Directive requires users to classify all surface waters according to the indicative ecological status, which gives a more holistic view of the assessment of the condition of a given ecosystem. The examined ecological state of surface water allows for appropriate preventive actions against the deterioration of environmental conditions. However, many countries, including Poland, still have problems with the precise interpretation of diatom indicators in lakes. Many countries are also in the process of introducing ecological indicators (e.g. diatom indicators) for the assessment of aquatic ecosystems. The human impact on the environment and climate change led to a progressive decline in the diversity of diatoms in lakes. The components of β diversity (eg species turnover or nesting) and the factors influencing changes in β diversity are still poorly understood, although diversity plays an important role in explaining ecological processes. Understanding the mechanisms responsible for changes in the components of β diversity (species nesting and turnover) along with gradients in environmental factors is currently a major problem in the biology and conservation of many species. Estimating the β diversity allows to measure the differences between the communities. It also contributes to the understanding of the different levels of impact of environmental conditions and water quality on aquatic communities.

The aim of the doctoral dissertation was to investigate the influence of selected environmental factors on the species diversity of diatoms and on the values of diatom indicators in the lakes of the Wigry National Park.

The study examined how selected environmental factors affect diatomaceous communities. It was analyzed **(in article 1: Diversity 2020)** how environmental differences between lakes a) disharmonic, b) harmonious with greater human impact on the environment and c) harmonious with more limited human impact are affecting diatom assembles. Significant differences were seen in the α Index of the Dominance Index. Overall, as study shows three groups are characterized by different environmental factors especially by pH, chloride and sulfate ions which is reflected by different dominance structure. The differences in the

diversity of β and the Diatom Index of Lakes (IOJ) and the reactions of diatoms to selected environmental factors were investigated **(in article 2: Water 2022).** The representativeness of the samples in estimating the ecological state of the lake was also examined. Results have proved that grouping of similar species decrease in the accuracy of the environmental assessment. Additionally, in article 3, precise taxonomic studies indicate and document how valuable and rich in rare species and protected are the lakes of the Wigry National Park **(in article 3: manuscript)**.

The work concerns the analysis of changes in the composition of diatom communities in environmental gradients, with particular emphasis on the diversity of α and β and with changes in the value of the Diatom Index of Lakes. The research results concern the differences in the diversity of α and β depending on anthropogenic changes in aquatic ecosystems. Environmental changes should be appropriately measured, especially in the most vulnerable freshwater ecosystems.

WSTĘP

Jeziora są ważnym elementem krajobrazu i istotnym biotopem dla unikatowych słodkowodnych biocenoz. Charakteryzują się dużą bioróżnorodnością gatunkową. Są wrażliwe na zmiany środowiskowe i klimatyczne (Ognjanova-Rumenova i inni 2019, Krztoń i inni 2019). Jeziora są też istotnym środowiskiem działalności człowieka, oraz są bardzo zagrożone antropopresją (Strayer i Findlay 2010). Szczególnie ważne jest monitorowanie ich obecnego stanu (Ognjanova-Rumenova i inni). Jeziorom zagrażają skutki bezpośrednie (np. wzbogacanie w składniki odżywcze, praktyki leśne i presja ze strony rolnictwa) oraz pośrednie (np. zmiany klimatyczne) (Ognjanova-Rumenova i inni 2019). W ciągu ostatnich kilkudziesięciu lat interdyscyplinarne badania wykazały rosnący wpływ zmian klimatycznych na skład chemiczny wód w jeziorach, co wiąże się z niezwykle istotnym spadkiem bioróżnorodności gatunkowej (Morandín-Ahuerma i inni 2019, Ognjanova-Rumenova i inni 2019).

Okrzemki (Bacillariophyta) należą do biowskaźników wykorzystywanych w ocenie cech przyrodniczych i antropogenicznych środowisk wodnych (Dam i inni 1994, Hofmann i inni 2011, Marra i inni 2018, Cristóbal i inni 2020). Okrzemki są uważane za jedną z najlepszych grup bioty wykorzystywanych do oceny statusu ekologicznego zbiorników wodnych (Dyrektywa 2000/60/WE). Różnorodność gatunkowa i rozmieszczenie okrzemek zależą od warunków środowiskowych, a struktura ich zespołów jest użytecznym bioindykatorem warunków środowiskowych (Stroemer i Smol 1999, Eliasz i Wojtal 2020, Eliasz i inni 2022). Są doceniane jako organizmy dobrze prognozujące stan środowiska (Van Dam 1982, Ochieng i inni 2022).

Zrozumienie mechanizmów wpływających na strukturę zbiorowisk okrzemkowych wzdłuż gradientów środowiskowych z perspektywy komponentów różnorodności β (wymiana gatunków i zagnieżdżenie) (Baselga 2010, Legendre i inni 2005) jest obecnie głównym zagadnieniem ekologii i ochronie gatunków (Wu i inni 2020). Wpływ antropopresji jest wyraźnie widoczny w ukształtowaniu ilościowym i jakościowym zespołów okrzemek (wskaźnik różnorodności α) (Eliasz i Wojtal 2020) i β (Eliasz i inni 2022). Składnik β różnorodności (wymiana gatunków lub ich zagnieżdżenie) i czynniki odpowiadające za ich funkcjonowanie są nadal słabo poznane, mimo że różnorodność odgrywa ważną rolę w wyjaśnianiu wielu procesów ekologicznych (Maloufi 2016). Obecnie coraz więcej uwagi badaczy skupia się na β różnorodności i jej składowych (wymiana gatunków i ich zagnieżdżenie). Różnorodność β zaproponowana przez schemat Baslega (2012) opiera się na macierzy obecności-braku taksonu. Można ją podzielić na składowe wymiany gatunków (czyli zastępowanie gatunków — jeden gatunek zastępuje inny bez zmiany bogactwa gatunkowego) i zagnieżdżenia gatunków (różnice w różnorodności wynikające z pojawienia się lub utraty gatunku). Inne podejście zaproponowano w analizie różnorodności β, w skali lokalnej (Legendre i De Cáceres 2013) dla macierzy z ilościowym wkładem gatunków (LCBD — lokalny wkład do różnorodności β). W indeksie LCBD również wyodrębniono zastępowanie (odpowiednik wymiany gatunków) i zagnieżdżanie (Legendre i De Cáceres 2013). Na wymianę gatunków ma wpływ oddziaływanie środowiska, konkurencja i wydarzenia historyczne (np. zlodowacenia, zmiany klimatu) (Specziár i inni 2018). Na zagnieżdżanie się gatunków ma wpływ zanikanie gatunków na danym terenie i inne procesy ekologiczne (np. wpływ człowieka, bariery fizyczne itp.) (Legendre 2014). Wszystkie te metody są przydatne w analizie zmian siedlisk i ochrony środowiska.

Ramowa Dyrektywa Wodna wymaga sklasyfikowania wszystkich wód powierzchniowych według ich stanu ekologicznego i daje bardziej kompleksowy wgląd w jakość wód danego obszaru (Nõges i inni 2006). Do oceny jakości wód jezior w Polsce (Picińska-Fałtynowicz i Błachuta 2010, Zgrundo i inni 2018) stosowane są jednorazowe w ciągu roku metody analizy elementów biologicznych makrofitów, ichtiofauny, planktonu, fitobentosu, makrozoobentosu. Jednak wiele krajów, w tym Polska ma problem z właściwym wdrożeniem wskaźników okrzemkowych dla jezior (Schaumburg i inni 2004, Poikane i inni 2016, Kelly i inni 2019, Bielczyńska 2015, Ciecierska i Kolada 2014, Wiech i inni 2018). Zmienność zbiorowisk okrzemek jest związana z sezonem (Elias i inni 2012, Hassan 2018), składem chemicznym wód i cechami fizycznymi zbiornika(Kelly i inni 2009). Reżim hydrologiczny, światło i konkurencja również wpływają na fitobentos okrzemkowy (Kelly i inni 2009, Rimet i inni 2015).

CEL BADAŃ

W badaniach podjęto problem wpływu wybranych czynników środowiskowych na różnorodność okrzemek oraz na wartości wskaźników okrzemkowych jezior Wigierskiego Parku Narodowego. Celem badań było ustalenie: *Jak czynniki środowiskowe wpływają na zbiorowiska okrzemkowe oraz na indeksy służące do opisu tych zbiorowisk (α i β różnorodność, IOJ)*?
W celu odpowiedzi na to pytanie założono przetestowanie następujących hipotez:

H1: α różnorodność okrzemek zmienia się wraz ze wzrostem wartości wybranych czynników fizyczno-chemicznych wód (artykuł nr 1: Eliasz-Kowalska, M., Wojtal, A. Z. **Limnological Characteristics and Diatom Dominants in Lakes of Northeastern Poland.** Diversity 2020, 12 (10), 374. https://doi.org/10.3390/d12100374. IF=3.029, MEiN=70)

H2: β różnorodność okrzemek zmienia się wraz ze wzrostem wartości wybranych czynników fizyczno-chemicznych wód (artykuł nr 2: Eliasz-Kowalska, M., Wojtal, A. Z., Barinova, S. **Influence of Selected Environmental Factors on Diatom β Diversity (Bacillariophyta) and the Value of Diatom Indices and Sampling Issues**. Water 2022, 14 (15), 2315. ttps://doi.org/10.3390/w14152315. IF=3.530, MEiN=100.)

H3: Indeks Okrzemkowy Jezior daje różne wyniki wraz ze wzrostem wartości wybranych czynników fizyczno-chemicznych wód, pory roku i miejsca poboru próby (**artykuł nr 2: Eliasz-Kowalska, M., Wojtal, A. Z., Barinova, S. Influence of Selected Environmental Factors on Diatom β Diversity (Bacillariophyta) and the Value of Diatom Indices and Sampling Issues**. Water 2022, 14 (15), 2315. ttps://doi.org/10.3390/w14152315. IF=3.530, MEiN=100.)

Prezentowana praca obejmuje trzy publikacje naukowe dotyczące badań terenowych laboratoryjnych i symulacji dotyczących okrzemkek obecnych w fitobentosie oraz wyselekcjonowanych indeksów służących do ich opisu w kontekście wybranych czynników środowiskowych.

TEREN BADAŃ

Do realizacji celów badawczych wybrano obszar w północno wschodniej Polsce - Wigierski Park Narodowy (WPN). Wigierski Park Narodowy jest terenem o wysokiej jeziorności i na małym obszarze można znaleźć różne typy limnologiczne jezior (np. dystroficzne, mezotroficzne, eutroficzne). Badania terenowe obejmowały próbki pobrane w sezonach wegetacyjnych 2015, 2016, 2017 i 2018. Obejmowały swoim zasięgiem 10 jezior (Fig. 1, Tab. 1) Białe Wigierskie, Wigry, Okrągłe, Krusznik, Muliczne, Białe Pierciańskie, Suchar Wielki, Suchar III, Wądołek i Wygorzele.

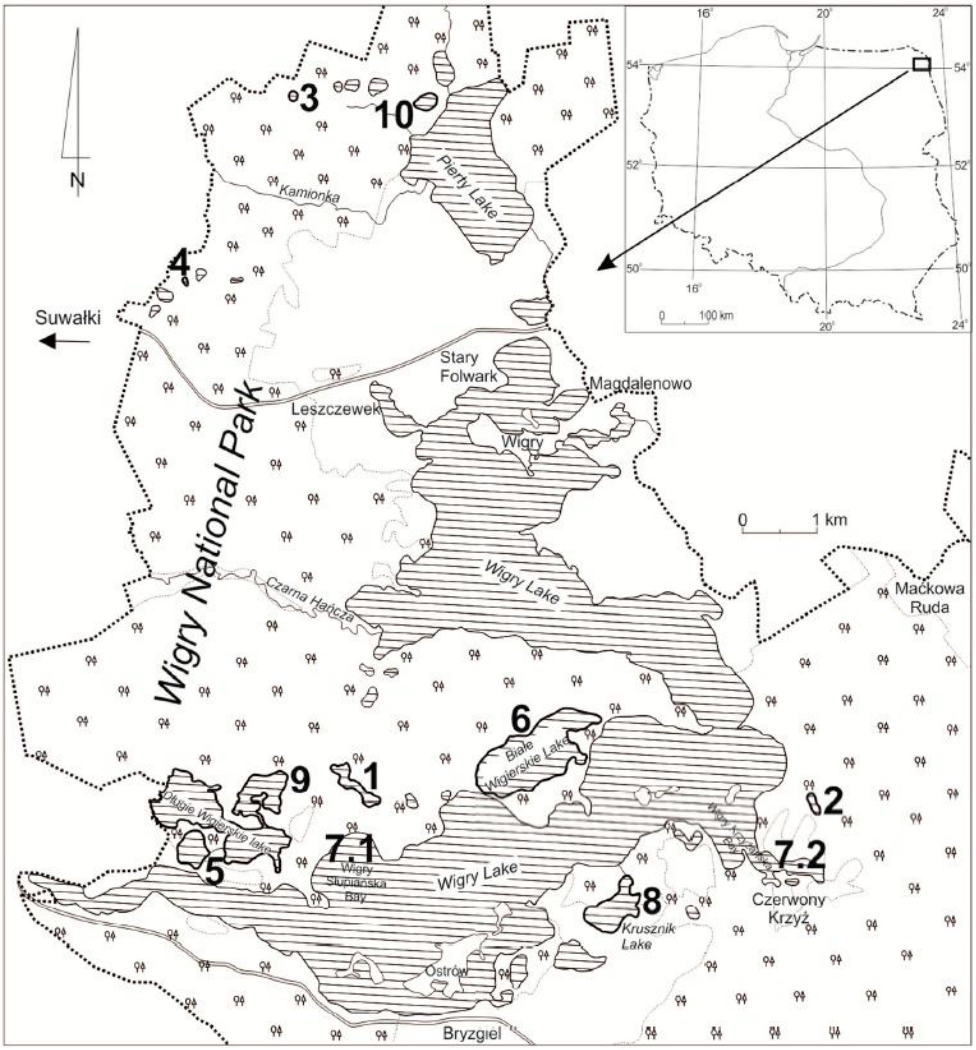

FIG . 1. Mapa terenu badań. Analizowane jeziora: 1—Suchar Wielki (SW), 2—Wygorzele (WYG), 3—Wądołek (WAD), 4—Suchar III (SIII), 5—Okrągłe (OK), 6—Białe Wigierskie (BW), Wigry (7.1—Słupiańska Bay (WS), 7.2— Krzyżańska Bay (WK), 8—Krusznik (K), 9—Muliczne (M), 10—Białe Pierciańskie (BP).

Nazwa Jeziora	Pow. jeziora [ha]	Gł. [m]	Dł. linii brzegowej [m]	Zlewnia bezpośrednia [ha]	Zlewnia [ha]	Rodzaj jeziora	Grupa jeziorna
Wigry	2163.3	73	63920	5159.8	45293.1	przepływowe	Wigierskie
Krusznik	26.7	18	2643	70.7	70.7	egzoreiczne	Wigierskie
Białe Wigierskie	99.9	34	5117	329.1	329.1	egzoreiczne	Wigierskie
Muliczne	24.1	11.3	3175	191.2	191.2	egzoreiczne	Wigierskie
Okrągłe	13.7	13	1459	28.5	906.8	przepływowe	Wigierskie
Białe Pierciańskie	6.9	24	1011	50.4	50.4	endoreiczne	Pierciańskie
Wygorzele	2	3	670	63.5	63.5	endoreiczne	Wigierskie
Suchar Wielki	8.44	9.6	2066	107.1	107.1	endoreiczne	Wigierskie
Wądołek	1.09	15	474	19.4	19.4	endoreiczne	Pierciańskie
Suchar III	0.44	4	320	32.2	32,2	egzoreiczne	Huciańskie

TAB. 1. Opis cech fizycznych jezior. Analizowane jeziora: BP – jez. Białe Pierciańskie, BW – jez. Białe Wigierskie, O- jez. Okrągłe, K – jez. Krusznik, M – jez. Muliczne, SIII – jez. Suchar III, SW – jez. Suchar Wielki, WYG – jez. Wygorzele, WAD – jez. Wądołek, W – jez. Wigry (za Górniak 2006).

Cały obszar WPN znajduje się pod wpływem klimatu umiarkowanego przejściowego między klimatem morskim a kontynentalnym (Andrzejczyk i Brzezicki 1995, Drzymulska i Zieliński 2014). W porównaniu z innymi pojezierzami Polski WPN znany jest z niskiej średniej rocznej temperatury (7,2°C) i wydłużonym czasem utrzymywania się pokrywy śnieżnej (Górniak 2020). Jeziora są pochodzenia lodowcowego i polodowcowego. Wszystkie są pozostałościami ostatniego okresu zlodowacenia, czyli zlodowacenia Weichsela (Górniak 2006). Wiele jezior stanowiło niegdyś część jednego większego akwenu późno plejstoceńskiego zwanego pre-Wigry. Niektóre badane jeziora: Białe Wigierskie, Okrągłe, Krusznik, Muliczne tworzą grupę polodowcowych zbiorników akumulacyjnych i separacyjnych. Jezioro Suchar III ma podobną genezę, związaną z procesem fragmentacji, w wyniku którego powstały mniejsze zbiorniki wód powierzchniowych (Górniak 2006). Jezioro polodowcowe powstałe syngenetycznie z wycofywaniem się lądolodu to z kolei jezioro Białe Pierciańskie, natomiast jezioro Suchar Wielki jest standardowym typem związanym z topnieniem brył martwego lodu (Górniak 2006). Powstałe w czasie zróżnicowanie chemiczne badanych jezior jest wynikiem naturalnych różnic geologicznych, a także odmiennych oddziaływań antropogenicznych na tym terenie. Największym badanym jeziorem jest jezioro właściwe Wigry (2163,3 ha), natomiast wszystkie inne akweny są znacznie mniejsze (powierzchnia poniżej 100 ha). W rzeczywistości połowa zajmuje tylko kilka hektarów powierzchni (Tab. 1). Jeziora Parku Narodowego zaliczane są do grupy Wigierskiej (Jeziora Wigry, Białe Wigierskie, Krusznik, Okrągłe, Muliczne, Suchar Wielki, Wygorzele), Huciańskiego (Jezioro Suchar III) i Pierciańskiego (Jeziora Wądołek i Białe Pierciańskie). Do grupy jezior harmonijnych należą: Białe Wigierskie, Białe Pierciańskie, Wigry, Krusznik, Muliczne i Okrągłe, za to do jezior dystroficznych należą: Suchar Wielki, Wądołek, Suchar III oraz Wygorzele. Jeziora są zazwyczaj połączone siecią mniejszych strumyków lub rzek, są to jeziora egzoreiczne (ciek wodny wypływa z jeziora) takie jak Krusznik, czy też jeziora przepływowe (ciek wodny wpływa i wypływa z jeziora) - Wigry. Wyjątkiem są tu jeziora endoreiczne, takie jak Jezioro Wygorzele, w których dopływ wody z zewnątrz najczęściej pochodzący z opadów na zlewnię bezpośrednią, jest równoważony przez parowanie.

METODY

Szczegóły metodyki omówiono w każdym z artykułów będących częścią niniejszej rozprawy. Łącznie przeanalizowano 92 próbki wody pod kątem ich właściwości fizycznych i chemicznych. Pobierano je wiosną (maj 2015, maj 2017, maj 2018), latem (wrzesień 2017, październik 2017) i jesienią (listopad 2016, wrzesień 2018). Wszystkie próbki pod kątem właściwości fizycznych i chemicznych pobrano na otwartej wodzie, przy czym wybrano porównywalne środowiska. Przewodność i pH mierzono za pomocą wieloparametrowej sondy YSI 6600 V2. Próbki wody do analiz chemicznych pobierano 20–30 cm pod powierzchnią jeziora przy użyciu butelek polietylenowych o pojemności 0,33 l i przechowywano w ciemności w temperaturze 4°C, aby ograniczyć zachodzące reakcje chemiczne. Analizy jonowe dotyczyły fosforanów, siarczanów, azotanów, fluorków, węglanów, chlorków i azotynów, a także sodu, litu, potasu, magnezu, amonu i wapnia. Pomiary laboratoryjne przewodnictwa i pH przeprowadzono na chromatografie jonowym Dionex w laboratorium Instytutu Ochrony Przyrody PAN. Pobrano peryfiton (z trzciny pospolitej *Phragmites australis* w jeziorach harmonijnych i *Carex spp.* (w jeziorach dysharmonijnych) z dziesięciu jezior (Fig. 1). Próbki okrzemek pobrano w tym samym czasie co próbki wody. W laboratorium próbki oczyszczono przez dodanie i ogrzanie w 37% H_2O_2. Reakcję zakończono przez dodanie $KMnO_4$ i HCl. Oczyszczony materiał okrzemkowy wysuszony na szkiełkach nakrywkowych osadzono w żywicy syntetycznej Naphrax®. Następnie materiał został przeanalizowany przy użyciu mikroskopu Nikon Eclipse–80i, licząc ok. 400 okazów. Do identyfikacji okrzemek użyto literatury specjalistycznej (Hofmann i inni 2011, Wojtal i inni 2011, Lange-Bertalot i Ulrich 2014, Van De Vijver 2014, Delgado i inni 2015, Lange-Bertalot i inni 2017, Kennedy i Allott 2017 oraz Heudre i inni 2019). Dominanty definiowane są w pracy jako gatunki występujące ze względną liczebnością powyżej 10%. Subdominanty to taksony o liczebności od 5 do 9,9%. Obliczono również wskaźniki okrzemkowe (IOJ indeks okrzemek dla jezior polskich) za pomocą programu dostarczonego przez GIOŚ (wersja 2010 i zaktualizowana z maja 2019). Analizowane macierze próbek wykorzystano do obliczenia zróżnicowania β — (1) całkowitego wskaźnika różnorodności Sørensena w parach z podziałem (wymiana gatunków i zagnieżdżenie) oraz (2) lokalny udział w zróżnicowaniu beta (LCBD) z podziałem również na wymianę gatunków i zagnieżdżenie.

Do danych chemicznych zastosowano hierarchiczną analizę skupień (HCA), aby zidentyfikować główne źródła zmienności w danych jeziorach i stworzyć klasyfikację analizowanych zbiorników wodnych. Analiza głównych składowych została wykorzystana do wizualizacji grupowania próbek zgodnie ze zmiennymi fizycznymi i chemicznymi. Połączenie HCA i PCA umożliwiło dokładniejszą interpretację zmienności występującej w chemii wody w jeziorach, a także zmiany charakteryzującej zespoły. Wskaźnik dominacji (za McNaughton i Wolf 1970) obliczono jako DI = p1 + p2, czyli sumę dwóch najwyższych wartości liczebności (%) w próbie. Zastosowano uogólnione modele liniowe z kategoryczną zmienną niezależną (grupa), aby zbadać różnice między grupami.

Aby określić ilościowo związek każdego składnika różnorodności β z czynnikami przestrzennymi i środowiskowymi, zastosowano regresję wielokrotną na macierzach odległości (Lichstein 2009, MRM). Zależność między wskaźnikami różnorodności β w macierzy odległości a wybranymi zmiennymi została zmodelowana przy użyciu algorytmu lasu losowego (RF) (Biau i Scornet 2016). Aby zbadać zmiany różnorodności β w wybranych czynnikach środowiskowych zbadano relacje między czynnikami środowiskowymi za pomocą modeli liniowych. Przeprowadzono analizy podziału zmienności, a także analizę RDA (analiza redundancji Van den Wollenberg 1977, Heggen 2012, Vilmi i inni 2015), aby sprawdzić, który ze wskaźników różnorodności β lepiej odpowiada chemicznym i fizycznym cechom wody.

Różnice między porami roku zostały przeanalizowane poprzez przeprowadzenie skalowania wielowymiarowego (NMDS, Kenkel i Orloci 1986) oraz PERMANOVA (permutacyjna analiza wariancji, Anderson 2017), która została wykonana w celu oceny różnic statystycznych między porami roku i jeziorami: w zbiorowiskach okrzemek oraz we wskaźnikach i ocenach opartych na indeksie okrzemek. Użyto RDA (Van den Wollenberg 1977, Heggen 2012, Vilmi i inni 2015), aby ocenić, który ze wskaźników okrzemkowych wyjaśniał więcej wariancji w zbiorze danych środowiskowych. Gatunki charakterystyczne dla każdego jeziora zostały określone przy użyciu funkcji IndVal (Zuur i inni 2007)

WYNIKI

W artykule nr 1 (Hipoteza 1; Diversity 2020) zbadano czy i jak zmieniają się zbiorowiska okrzemek w stosunku do zmian wybranych czynników środowiskowych, ze szczególnym uwzględnieniem α różnorodności. Jako szczególnie różnicujące czynniki środowiskowe zostały wybrane pH, jony siarczanowe (SO_4^{2-}) i chlorkowe (Cl^-). Są one najsilniejszymi zmiennymi objaśniającymi badanego wskaźnika α, czyli wskaźnika dominacji. Do analiz wykorzystano PCA (Principal Component Analysis (Ringnér 2008) i HCA (Hierarchical Cluster Analysis (Boggero i inni 2019) w celu oceny, które czynniki środowiskowe najsilniej odpowiadają za zmienność oraz w celu podzielenia jezior na grupy o podobnych właściwościach fizycznych i chemicznych wody, grupowanie zostało zbadane współczynnikiem Silhouette [Nidheesh i inni 2020) oraz siłą predykcji (prediction strength (Tibshirani i Walther 2005)], potwierdzając skuteczność podziału na grupy. W badaniu pokazano, że badane jeziora można podzielić na 3 grupy, charakteryzujące się w szczególności różnicami w pH oraz zawartości jonów siarczanowych i chlorkowych. Grupy te nazwano: a) dysharmonijnymi, b) harmonijnymi z większą antropopresją i c) harmonijnymi z bardziej ograniczonym wpływem człowieka ze względu na ich charakterystykę. Użyto GLM (Generalized Linear Models) w celu oceny istotności różnic pomiędzy grupami. Różnice te okazały się istotne statystycznie (Fig. 2).

W grupie a) dysharmonijnej najbardziej charakterystycznym dominantem (Suplement A) była *Tabellaria flocculosa* (Roth) Kützing 1844. Dominantami były również takie gatunki jak *Eunotia mucophila* (Lange-Bertalot, Nörpel-Schempp i Alles), Lange-Bertalot 2007, *E. rhomboidea* Hustedt 1950, *E. genuflexa* Nörpel-Schempp 1996*, Stauroforma exiguiformis* (Lange-Bertalot), R.J.Flower, V.J.Jones i Round 1996, *Kobayasiella subtillissima* (Cleve) Lange-Bertalot 1999 i *Nitzschia gracilis* Hantzsch 1860. Druga grupa (Suplement A) b) była zdominowana wyłącznie przez *Achnanthidium minutissimum* (Kützing) Czarnecki 1994, *Fragilaria subconstricta* Østrup 1910 i *Encyonopsis microcephala* (Grunow) Krammer 1997.

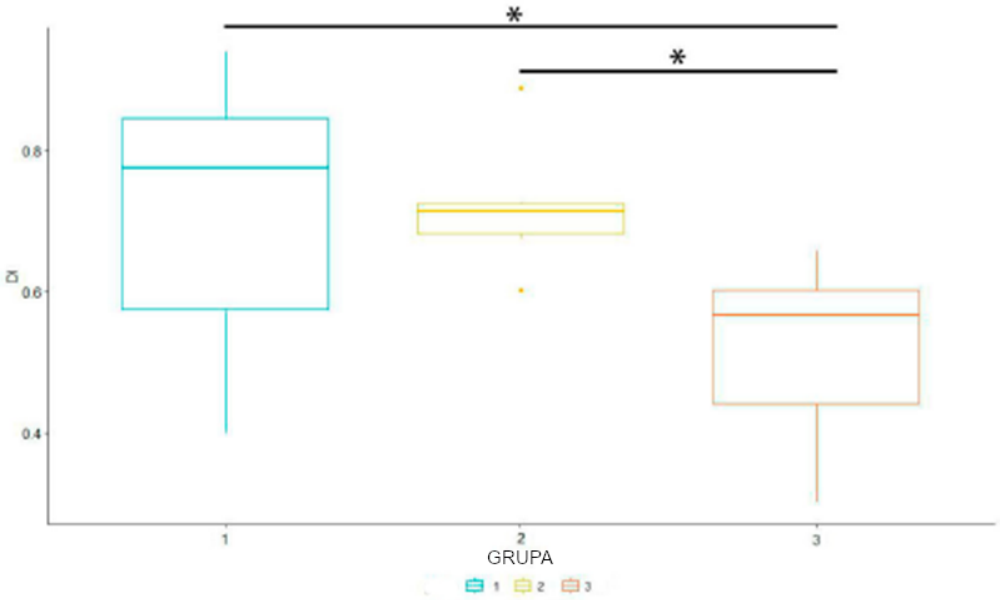

FIG. 2. GLM (Analiza ogólnego modelu liniowego) dla badanych grup a) dysharmonijnymi, b) harmonijnymi z większą antropopresją i c) harmonijnymi z bardziej ograniczonym wpływem człowieka, DI – wskaźnik dominacji, gwiazdką zaznaczone różnice istotne statystycznie.

Achnanthidium minutissimum jest obserwowane w szerokim zakresie właściwości fizycznych i chemicznych wód, podczas gdy *Fragilaria subconstricta* występuje w mezotroficznych i lekko alkalicznych wodach. *Encyonopsis microcephala* występuje w oligotroficznych do lekko eutroficznych wodach (Heudre i inni 2019, Lange-Bertalot i inni 2017). Wymienione gatunki są bardziej tolerancyjne na antropopresję i większe stężenia nutrietów niż gatunki z grupy trzeciej - c).

Trzecia grupa c) posiadała następujące gatunki dominanujące (Suplement A): *Achnanthidium minutissimum, Encyonopsis microcephala,* oraz dodatkowo *Brachysira microcephala* (Grunow) Compère 1986, *Encyonopsis cesatii* (Rabenhorst) Krammer 1997, *Eunotia arcubus* Nörpel i Lange-Bertalot 1993, *Delicatophycus delicatulus* (Kützing) M.J.Wynne 2019 i *Staurosirella pinnata* (Ehrenberg) D.M.Williams i Round 1988. Wymienione gatunki wskazują na oligotroficzne do mezotroficznych właściwości wody oraz wysoką zawartość węglanów wapnia (Lange-Bertalot i inni 2017). Gatunki po raz pierwszy stwierdzone dla WPN w tym artykule to *Delicatophycus delicatulus, Encyonopsis cesatii, Fragilaria subconstricta* i *Eunotia arcubus.* Różnice w strukturze dominacji były związane z niską różnorodnością gatunkową oraz niższą równomiernością gatunków. Przy spadku pH zaobserwowano silniejszą strukturę dominacji, co jest zgodne z obserwacjami innych naukowców (Witkowski i inni 2011). W takim środowisku następuje obniżenie tolerancji gatunków i są one bardziej wrażliwe na inne stresory. Dla jezior z grupy harmonijnej o niższej antropopresji (c) zaobserwowano niższą dominację gatunków, większą różnorodność i równomierność gatunków. Van Dam (1994) opisywał iż taki skok bioróżnorodności może być spowodowany umiarkowanymi zaburzeniami w środowisku. Wychodząc z założenia, że oczekiwana naturalna struktura zbiorowisk okrzemkowych w jeziorach składa się z jednego lub dwóch silnych dominantów, kilku subdominantów i rzadkich gatunków w mniejszości, to wprowadzenie umiarkowanych zaburzeń, np. suszy (Calapez i inni 2014), jest związane z przebudową zbiorowiska z dominantów przystosowanych do warunków sprzed zaburzenia i dominantów dostosowanych do zaburzenia. Skutkuje to tymczasowym zwiększeniem różnorodności. Silniejsze zaburzenia w środowisku powodują homogenizację środowiska i silny spadek bioróżnorodności. Tego typu obserwacje są zgodne z Hipotezą Umiarkowanych Zaburzeń (Intermediate Disturbance Hypothesis) (Connell 1978). Jednak tego typu struktura zbiorowisk okrzemek w przypadku analizowanych w badaniach jezior powtarza się w różnych sezonach, co budzi wątpliwości, czy hipoteza ma tu również zastosowanie. W szczególności niejasnym w tym kontekście pozostaje wyższa bioróżnorodność w zbiorowiskach obserwowana dla jezior szczególnie chronionych i oddalonych od źródeł zanieczyszczeń, jak np. jezioro Białe Pierciańskie. W ostatnich latach IDH było przedmiotem intensywnej debaty (np. Lengyel 2016, Fox 2013, Sheil 2013). Antropopresja (jak w grupie b) jest silnym stresorem zmniejszającym bioróżnorodność (Pandey 2017). Następuje wówczas przebudowa zbiorowisk w kierunku gatunków mniej wrażliwych na wysokie stężenia nutrientów (Schneider i inni 2019). W analizowanym przypadku zmiany w strukturze dominacji okrzemek mogą być związane ze zmianami fizycznymi i chemicznymi właściwości wód w jeziorach w kierunku bardziej powszechnym w czasie ewolucji okrzemek (tu np. bardziej ubogiej w elektrolity, czy też przeskok z wód o niskim pH do circumneutralnych). W takich środowiskach dostępnych jest więcej gatunków wyspecjalizowanych ze względu na dłuższy czas ewolucji stąd też bioróżnorodność jest wyższa, a dominacja niższa (Taylor i inni 1990, Pither i inni 2005), co jest zgodne z Hipotezą Puli Gatunków (Species Pool Hypothesis). Dodatkowo opublikowane wcześniej badania analizowanych terenów wskazują, iż najczęstszy rodzaj zbiorników charakteryzował się względnie wysokim pH, stosunkowo wysoką mineralizacją i oligotrofią (Zawisza i Szeroczyńska 2007, Witkowski i inni 2009). Zmiany w różnorodności okrzemek zgodne z Hipotezą Puli Gatunków (Species Pool

Hypothesis) były potwierdzane przez naukowców dla innych środowisk (Pither i inni 2005). Próby o wysokiej dominacji były reprezentowane głównie przez generalistów, gatunki wyspecjalizowane były obecne w mniejszości. Obserwacje te są zgodne z Soininen i Heino (2007). Niezgodne z Soininen i Heino (2007) dane otrzymano natomiast dla Grupy c, która była bardziej zróżnicowana. Była również reprezentowana głównie przez generalistów i w drugiej kolejności przez specjalistów, pomimo innego stosunku względem siebie niż w pierwszym przypadku. Uzyskany wynik natomiast jest zgodny z analizami dla jezior (Chen i inni 2016, MacDougall i inni 2017).

W artykule nr 2 (Hipoteza 2; Water 2022) analizowano czy i jak zmieniają się zbiorowiska okrzemek w stosunku do zmian wybranych czynników środowiskowych, ze szczególnym uwzględnieniem β różnorodności. Jako szczególnie różnicujące faktory środowiskowe zostały wykazane jony siarczanowe, azotanowe, fosforowe, amonowe i wapnia. Wszystkie wskaźniki β różnorodności istotnie statystycznie opisywały zmienność chemiczną fizyczną wód. Do analiz wykorzystano modele Liniowe (Linear Model) i Losowych Lasów Decyzyjny (Random Forest Model (Biau i Scornet 2016) (Tab. 2) wraz z metodami wyjaśniającymi – permutacyjną oceną cech [rfpimp – Random Forest Permutational Feature Importances (Altmann i inni 2010)] i średnim obniżeniem precyzji [eli5 – Mean Decrease Accuracy (Gómez-Ramírez i inni 2020), Podział Wariancji (Variation Partitioning), Analizę Redundancji (RDA – Redundancy Analysis (Van den Wollenberg 1977) oraz Regresję wielokrotną na Macierzach Odległości (Multiple Regression on Distance Matrices (MDM) (Lichstein 2006)]. Wykazano, że β różnorodność na badanych terenach składała się głównie z wymiany gatunków (średnia 81%, mediana 85%), zagnieżdżenie gatunków odpowiadało za średnio 19% wariancji (mediana 15%). Lokalny Udział w β różnorodności (Local Contribution to β Diversity – LCBD) również składał się głównie z wymiany gatunków (średnia 88% i mediana 92%). Wskaźniki β były silnie skorelowane z czynnikami środowiskowymi, najsilniejszymi predyktorami wskaźników β różnorodności Sørensena i Lokalnego Udziału w β różnorodności były jony siarczanowe i azotanowe, oba będące indykatorami eutrofizacji wody i zwiększonej antropopresji (Bennion i inni 2010). Zgodnie z opisanymi mechanizmami (Soininen 2014), w przypadku opisywanych badań, wymiana gatunków odgrywała największą rolę, co oznacza, że wpływ lokalnych czynników cech środowiskowych był najsilniejszy. Większe stężenie oraz różnice w jonach azotanowych i siarczanowych spowodowało większą wymianę gatunków oraz większe zagnieżdżenie gatunków. Badania pokazały również, iż dla jezior wskaźniki β różnorodności Sørensena dużo lepiej wyjaśniają właściwościowości fizyczne i chemiczne wód niż Lokalny Udział w β różnorodności (Local Contribution to β Diversity – LCBD). Wyraźna dominacja gatunków w jeziorach obniżała wartości dla Lokalnego Udziału w β różnorodności i siłę wyjaśniającą, podczas gdy wskaźniki Sørensena (SCBD) oparte na macierzy obecności-braku, precyzyjniej opisywały środowisko. Gatunki, które miały największy wpływ na β różnorodność *to Achnanthidium affinis, Brachysira neoexilis* i *Cymbella affiniformis*. Te gatunki były relatywnie często spotykane, jednak nie były najbardziej liczne. Za to najczęściej spotykane gatunki, obecne w większości próbek, miały niewielki wpływ na β różnorodność oraz wpływały negatywnie na LCBD. Analizy biorące pod uwagę macierz obecności-braku gatunków (SCBD) obniżały wagę najliczniejszych gatunków co pozwoliło na zwiększenie mocy wyjaśniającej wskaźników na nich opartych.

Analiza	Funkcja	LCBD całkowite	LCBD wymiana gatunków	LCBD zagnieżdżenie	Beta Sørensen całkowity	Beta Sørensen wymiana gatunków	Beta Sørensen zagnieżdżenie
1. RDA	Model variance constrained	0.3763	0.4172	0.2580	1.056	0.3421	0.716
	Model variance uncontrained	4.6237	4.5828	4.742	3.944	4.6579	4.284
	Model p	0.011*	0.006**	0.048*	0.001**	0.017*	0.001**
	Axis p	0.008**	0.006**	0.048*	0.002**	0.012*	0.002**
2. Podział wariancji	Adj $R^2 SO_4^{2-}$	0.013	0.01842	-0.02015	0.16979	-0.02171	0.33897
	Adj R^2 NO_3^-	0.00721	-0.01534	0.09121	0.45749	0.13798	0.20454
	Adj $R^2 PO_4^{3-}$	0.04678	0.03172	0.01942	0.00153	-0.02019	0.00510
	Adj $R^2 NH_4^+$	-0.01761	-0.01066	-0.01324	0.04875	0.03053	-0.01343
3. MRM	p SO_4^{2-}	-	-	-	-	-	-
	p NO_3^-	0.001**	0.001**	-	0.004**	0.020*	-
	p Ca^{2+}	-	-	-	0.001**		0.001**
	p Cl^-					0.001**	-
	p PO_4^{3-}	0.032*	0.018*	-	-	-	-
	R^2	0.1085	0.1124	Nie znacząca	0.2041	0.05953	0.1297
4. Las losowy (funkcja permutation_importances)	SO_4^{2-}	0.305	0.232	0.128	0.419	0.241	0.140
	NO_3^-	0.266	0.201	0.122	0.229	0.236	0.293
	Ca^{2+}	0.130	0.146	0.562	0.403	0.176	0.400

	Cl^-	0.210	0.278	0.121	0.079	0.212	0.150
	NH_4^+	0.067	0.097	0.189	0.195	0.125	0.167
	PO_4^{3-}	0.083	0.106	0.083	0.062	0.104	0.098
	R^2	0.83	0.85	0.89	0.88	0.82	0.81
4. Las losowy (funkcja eli5)	SO_4^{2-}	0.299	0.241	0.129	0.409	0.251	0.123
	NO_3^-	0.236	0.193	0.135	0.245	0.263	0.284
	Ca^{2+}	0.132	0.159	0.536	0.351	0.169	0.371
	Cl^-	0.210	0.320	0.147	0.0756	0.223	0.166
	NH_4^+	0.071	0.104	0.191	0.160	0.143	0.167
	PO_4^{3-}	0.098	0.148	0.096	0.055	0.100	0.91
	OOB score	0.52	0.42	0.55	0.44	0.31	0.28
5. Model liniowy	p SO_4^{2-}	0.0020**	0.000811***	-	0.0311*	-	0.00000005***
	p NO_3^-	0.000549***	0.002330**	0.0204*	0.00000126***	0.00541**	0.00000126***
	p Ca^{2+}	0.000284***	0.000240***	-	-	-	0.0000852***
	p Cl^-	-	-	-	-	-	-
	p PO_4^{3-}	-	-	-	-	-	-
	p NH_4^+	0.0399*	0.044987*	-	-	-	0.0123*
	Adj R^2	0.26	0.257	0.0921	0.4992	0.138	0.597

TAB. 2. Modele dla różnorodności β. 1: Analiza redundancji (RDA) dla wybranego wskaźnika zróżnicowania β, 2: Podział wariancji i skorygowany R^2 dla analizowanych czynników. 3: Regresja wielokrotna na macierzach odległości (MRM), 4: Las losowy i opis modelu, 5. Model liniowy; Adj – skorygowany; R^2 — współczynnik determinacji; p — prawdopodobieństwo; *—p < 0,05; **—p < 0,01; ***—p < 0,001.

W artykule nr 2 (Water 2022) badano również **hipotezę 3** gdzie analizowano jak reprezentatywna jest próbka pobrana do analiz zbiorowiska z różnych punktów na jeziorze. W badaniach wykazano jak różnice w czasie i miejscu poboru próby rzutują na wyniki a) oceny zbiorowiska, b) Indeksu Okrzemkowego Jezior w wersji 2010 i 2019 jak i c) Oceny Jakości Ekologicznej Wody na podstawie IOJ. W tym celu zastosowano dwukierunkowe podejście. Przy użyciu PERMANOVA oceniano (Permutational Analysis of Variance) (Anderson 2017) czy a) występuje różnica w badanych poziomach, b) pomiędzy sezonami i różnymi jeziorami. Wygenerowano również próbki przy użyciu symulacji Monte Carlo (Heggen i inni 2012, Hassan 2018, Riato 2020). Następnie oceniano reprezentatywność próbek rzeczywistych (nie generowanych) porównując wyniki na badanych poziomach a), b) i c). Próbki dla poziomu a) (Fig. 3) oceniane były jako wysoce różne, gdy mieściły się w ostatnich 5% odległości od centroidów, dla poziomów b) i c) kiedy mieściły się w ostatnich 2.5% wartości po obu stronach histogramu. Oprócz tego oceniano przy pomocy RDA siłę predykcji na poziomie b) Indeksu Okrzemkowego Jezior. Badania wykazały, że występują różnice istotne statystycznie pomiędzy zbiorowiskami (poziom a) w różnych porach roku oraz w badanych jeziorach (Fig. 4). Wszystkie badane pory roku i jeziora wykazywały się różnicą istotną statystycznie. Różnice zaobserwowano również dla poziomu b) Indeksu Okrzemkowego Jezior, co oznacza, że Indeks Okrzemkowy był zależny od jeziora i pory roku. Na poziomie c) praktycznie różnice pomiędzy jeziorami i sezonami nie występowały w szczególności dla wersji z uproszczonym procesem identyfikacji gatunkowej. RDA wykazało istotnie statystyczną predykcję dla wcześniejszej metody oceny Indeksu Okrzemkowego Jezior oraz nieistotną statystycznie predykcję dla metody z uproszczonym procesem identyfikacji gatunkowej. Wysoce różne próbki po względem poziomu a) to aż 79% wszystkich próbek. Wynikało to z wyraźnej dominacji gatunków i dużej zmienności gatunków towarzyszących. 69% próbek rzeczywistych (nie generowanych) było istotnie różnych pod względem poziomu b) co oznacza, że różnice w Indeksie Okrzemkowym Jezior pomiędzy stanowiskami występowały często i były duże. Różnice te jednak znikały na poziomie c) przy ocenie stanu środowiska, zgodnie z wynikami z Polski (Bielczyńska 2015, Ciecierska i Kolada 2014, Wiech i Marinkiewicz-Mykitta 2018).

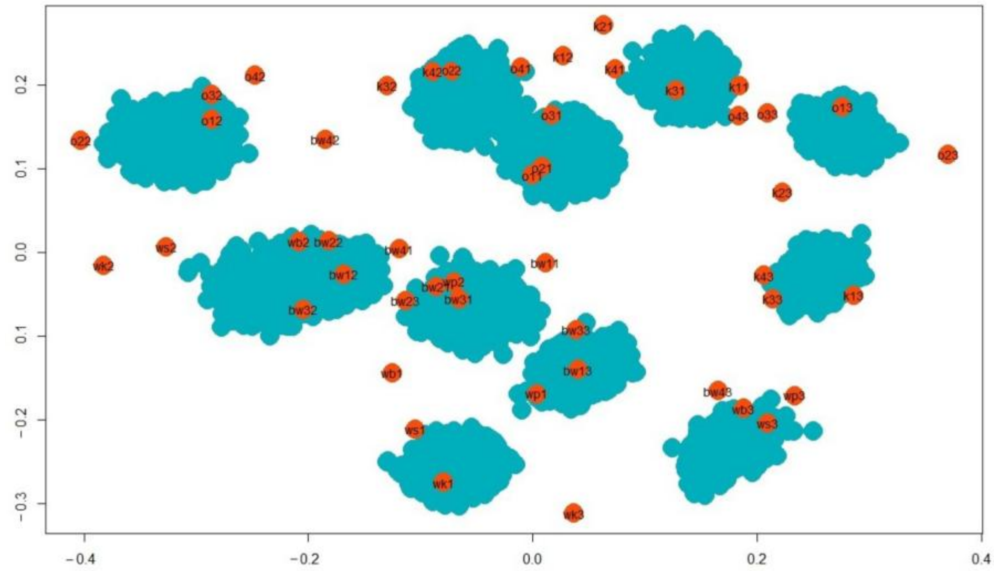

FIG. 3. Skalowanie wielowymiarowe dla symulowanych zbiorowisk (na niebiesko) i rzeczywistych próbek. Rysunek poglądowy, porównanie jezior między sobą.

Ocena środowiska przy pomocy IOJ bardzo rzadko daje wyniki różne od bardzo dobrej i dobrej, i jeszcze rzadziej IOJ odpowiada za końcową ocenę środowiska (zgodnie z zasadą one-out-all-out, Picińska-Fałtynowicz i Soszka 2011- jeden wskaźnik wskazuje gorszy stan, tzn. że ocena końcowa będzie od niego zależna). Wyniki otrzymanych analiz potwierdzają problem z Indeksem Okrzemkowym Jezior. Konieczne są zmiany w samym sposobie obliczania indeksu, tak żeby zwiększyć jego moc predykcyjną dla jezior. Z punktu widzenia ograniczeń sprzętowych i pracowniczych jest niemożliwe w tej chwili zwiększenie liczby pobieranych prób w sezonie i w badanym jeziorze (King 2006, Prygiel i inni 2002 Zgrundo et al. 2018 oraz Kelly i inni 2009). Niepewność związana z poborem prób (miejsce poboru i termin poboru) jest ograniczana wieloletnimi badaniami nad danym jeziorem.

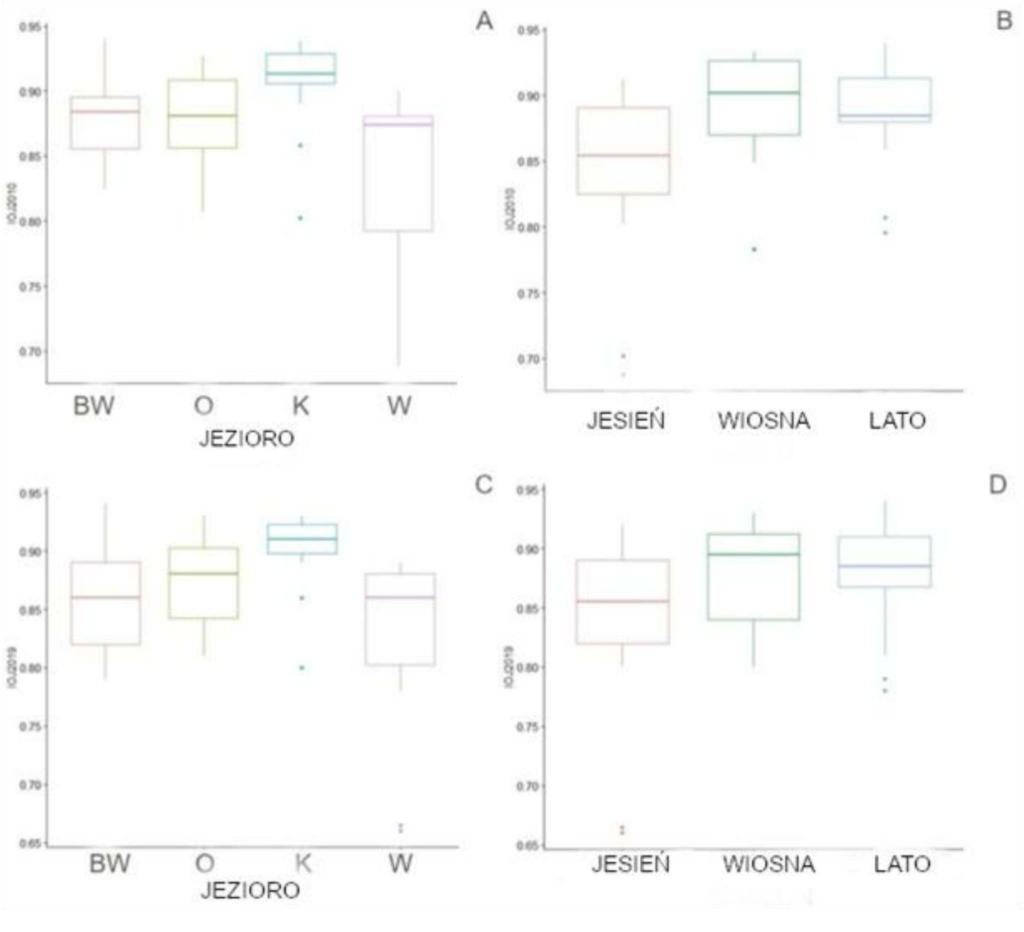

FIG. 4. Analiza ogólnego modelu liniowego (GLM) dla grup — jeziora (A, B) i pory roku (C, D); IOJ 2010 (A, C), IOJ 2019 (B, D). O – jezioro Okrągłe; BW – Jezioro Białe Wigierskie; K — jezioro Krusznik; W – jezioro Wigry.

WNIOSKI

Przeprowadzone badania, pozwoliły odpowiedzieć na pytanie: *Jak wybrane czynniki środowiskowe wpływają na zbiorowiska okrzemkowe oraz na indeksy okrzemkowe służące do opisu tych zbiorowisk (α i β różnorodność, IOJ)?*

Okrzemki są jednymi z najlepszych indykatorów jakości wody. Lepsze poznanie mechanizmów związanych z regulacją zbiorowisk oraz indeksów służących do opisu tych zbiorowisk jest ważne zarówno w aspekcie poznawczym jak i z punktu widzenia praktycznego użytkowania indeksów okrzemkowych i bioróżnorodności α i β do oceny jakości środowiska. W pracy zastosowano nowatorskie podejście do oceny indeksów wykorzystując symulację zbiorowisk i ocenę reprezentatywności próbek. Dzięki temu możliwa była ocena nie tylko same zbiorowiska, ale również wskaźniki obliczane z pojedynczej próbki i z grupy próbek.

Wyniki badań potwierdziły, że różnorodność α okrzemek rośnie w środowiskach ubogich w elektrolity o niższej antropopresji. Jest to zgodne z Hipotezą Puli Gatunków (Species Pool Hypothesis) (**Praca nr 1**: Eliasz i Wojtal 2020).

Wykazano, że β różnorodność na badanych terenach składała się głownie z komponentu - wymiany gatunków, w mniejszym stopniu z zagnieżdżenia (**Praca nr 2**: Eliasz i inni 2022). Przeprowadzone analizy wskazują, że dla jezior WPN wskaźniki β różnorodności Sørensena lepiej wyjaśniają właściwości fizyczne i chemiczne wód niż Lokalny Udział w β różnorodności (Local Contribution to β Diversity – LCBD). Wysoka dominacja gatunków w jeziorach (powyżej 70% dominantów – gatunków występujących w liczebności powyżej 10%) wpływała na obniżenie wartości LCBD (Lokalnego Udziału w β różnorodności) i na obniżenie siły wyjaśniającej, podczas gdy wskaźniki Sørensena oparte na macierzy obecności-braku, wiarygodniej opisywały środowisko. Jony siarczanowe i azotanowe, oba będące indykatorami eutrofizacji wody i zwiększonej antropopresji, były najsilniej skolerowane ze wskaźnikami β różnorodności Sørensena i Lokalnym Udziałem w β różnorodności.

Badania terenowe pokazały, że gatunki o dużej dominacji (powyżej 10%) w jeziorach WPN obniżają wiarygodność wskaźnikową IOJ oraz wskaźników Lokalnego Udziału w β różnorodności) (**Praca nr 2**: Eliasz i inni 2022). Wyniki te wskazują na odrębny kierunek badań dla lokalnych analiz β różnorodności dla jezior, niż polecany w dotychczasowych badaniach (zastosowanie Lokalnego Udziału (LCBD) w β różnorodności w badaniach na mniejszym terenie).

Połączenie badań terenowych oraz symulacja próbek i analiza wskaźników na podstawie próbek rzeczywistych i generowanych komputerowo jest oryginalnym rozwiązaniem weryfikacji hipotezy 3.

Nowym zagadnieniem poruszonym w moich badaniach była ocena zależności zbiorowisk na różnych poziomach oceny środowiska od czynników fizyczno chemicznych wód i porównanie ich z symulacjami zbiorowisk okrzemkowych. Wyniki pracy wykazały różnice w wynikach na poziomie zbiorowiska i oceny Indeksu Okrzemkowego Jezior, różnice te znikały przy ocenie stanu środowiska. Dane te mają wartość użytkową i są wskazówką do dalszej poprawy Indeksu Okrzemkowego Jezior.

PODSUMOWANIE

Przeprowadzone badania poszerzają wiedzę dotyczącą mechanizmów odpowiedzialnych za organizację zbiorowisk. Analizy pokazały iż pH, jony siarczanowe, azotanowe i chlorkowe są najsilniejszymi predykatorami badanych wskaźników α i β. Dla przykładowego wskaźnika α – Wskaźnika Dominacji, zależności te nie są liniowe. Mimo różnic pomiędzy wynikami indeksu okrzemkowego dla różnych stanowisk na jeziorze, jak i różnic pomiędzy porami roku, zwiększenie liczby poborów i inne zmiany metodyczne (przykładowo składanie gatunków w łatwiejsze do oznaczenia grupy) nie są wystarczającą metodą poprawy Indeksu Okrzemkowego Jezior.

Wyniki tej pracy można wykorzystać do poprawy wskaźników okrzemkowych dla jezior oraz przez kraje wdrażające oceny ekologiczne wód jezior.

LITERATURA

Altmann, A., Toloşi, L., Sander, O., Lengauer, T. Permutation Importance: A Corrected Feature Importance Measure. *Bioinformatics* **2010**, *26* (10), 1340–1347. https://doi.org/10.1093/bioinformatics/btq134.

Anderson, Marti J. Permutational multivariate analysis of variance (PERMANOVA). *Wiley statsref: statistics reference online* **2014,** 1-15.

Andrzejczyk, T., Brzeziecki, B. The Structure and Dynamics of Old-Growth Pinus Sylvestris (L.) Stands in the Wigry National Park, North-Eastern Poland. *Vegetatio* **1995**, *117* (1), 81–94. https://doi.org/10.1007/BF00033261.

Baselga, A. Partitioning the Turnover and Nestedness Components of Beta Diversity: Partitioning Beta Diversity. *Glob. Ecol. Biogeogr.* **2010**, *19* (1), 134–143. https://doi.org/10.1111/j.1466-8238.2009.00490.x.

Baselga, A., Orme, C. D. L. Betapart : An R Package for the Study of Beta Diversity: *Betapart Package*. *Methods in Ecology and Evolution* **2012**, *3* (5), 808–812. https://doi.org/10.1111/j.2041-210X.2012.00224.x.

Bennion, H., Sayer, C. D., Tibby, J., Carrick, H. J. Diatoms as Indicators of Environmental Change in Shallow Lakes. In *The Diatoms: Applications for the Environmental and Earth Sciences*, Cambridge University Press: Cambridge, **2010**.

Biau, G., Scornet, E. *A Random Forest Guided Tour*. *TEST* **2016**, *25*, 197–227.

Bielczyńska, A. Bioindication on the Basis of Benthic Diatoms: Advantages and Disadvantages of the Polish Phytobenthos Lake Assessment Method (IOJ—The Diatom Index for Lakes)/Bioindykacja Na Podstawie Okrzemek Bentosowych: Mocne i Słabe Strony Polskiej Metody Oceny Jezior Na Podstawie Fitobentosu (IOJ—Indeks Okrzemkowy Jezior). *Ochr. Śr. Zasobów Nat.* **2015**, *26*, 48–55.

Birk, S., Bonne, W., Borja, A., Brucet, S., Courrat, A., Poikane, S., Solimini, A., van de Bund, W., Zampoukas, N., Hering, D. Three Hundred Ways to Assess Europe's Surface Waters: An Almost Complete Overview of Biological Methods to Implement the Water Framework Directive. *Ecol. Indic.* **2012**, 18, 31–41. https://doi.org/10.1016/j.ecolind.2011.10.009.

Boggero, A., Zaupa S., Musazzi S., Rogora M., Dumnicka E., Lami A. Environmental factors as drivers for macroinvertebrate and diatom diversity in Alpine lakes: New insights from the Stelvio National Park (Italy). *Journal of limnology.* **2019,** 78: 147–162. DOI: 10.4081/jlimnol.

Calapez, A. R., C. L. Elias, S. F. Almeida& M. J. Feio. Extreme drought effects and recovery patterns in the benthic communities of temperate streams. *Limnetica.* **2014,** 33: 281–296. DOI: 10.23818/limn.33.22

Chen, W., Wang, Q., Xia, S., Pei, G. Comparative responses of dominant and subordinate periphytic diatom species to environmental gradients in Donghu Lake, China. *International Review of Hydrobiology*, **2016**, 101(5-6), 182-192.
DOI: 10.1002/iroh.201601845

Ciecierska, H., Kolada, A. ESMI: a macrophyte index for assessing the ecological status of lakes. *Environmental monitoring and assessment,* **2014,** *186*(9), 5501-5517.
DOI: 10.1007/s10661-014-3799-1

Connell, J. H. Diversity in tropical rain forests and coral reefs. *Science*, **1978**, 199: 1302–1310. DOI: 10.1126/science.199.4335.1302

Cristóbal, G., Blanco, S., Bueno, G. (Eds.) Modern Trends in Diatom Identification: Fundamentals and Applications, *Developments in Applied Phycology*, Springer: Cham, Switzerland, **2020**.

Dam, H., Mertens, A., Sinkeldam, J. A Coded Checklist and Ecological Indicator Values of Freshwater Diatoms from The Netherlands. *Neth. J. Aquat. Ecol*. **1994**, 28 (1), 117–133. https://doi.org/10.1007/BF02334251.

Delgado, C., Novais, M. H., Blanco, S., F.P. de Almeida, S. Examination and Comparison of Fragilaria Candidagilae Sp. Nov. with Type Material of Fragilaria Recapitellata, F. Capucina, F. Perminuta, F. Intermedia and F. Neointermedia (Fragilariales, Bacillariophyceae). *Phytotaxa* **2015**, *231* (1), 1.
https://doi.org/10.11646/phytotaxa.231.1.1.

Drzymulska, D., Zieliński P. Phases and interruptions in postglacial development of humic lake margin (Lake Suchar Wielki, NE Poland). *Limnological Review*, **2014**, 14: 13–20.
DOI: 10.2478/limre-2014-0002

Elias, C. L., Vieira, N., Feio, M. J., Almeida, S. F. P. *Can Season Interfere with Diatom Ecological Quality Assessment? Hydrobiologia*, **2012**, 695 (1), 223–232.
https://doi.org/10.1007/s10750-012-1196-8.

Eliasz-Kowalska, M., Wojtal, A. Z. Limnological Characteristics and Diatom Dominants in Lakes of Northeastern Poland. *Diversity* **2020**, *12* (10), 374.
https://doi.org/10.3390/d12100374.

Eliasz-Kowalska, M., Wojtal, A. Z., Barinova, S. Influence of Selected Environmental Factors on Diatom β Diversity (Bacillariophyta) and the Value of Diatom Indices and Sampling Issues. *Water* **2022**, *14* (15), 2315.
https://doi.org/10.3390/w14152315.

Fox, J. W. The intermediate disturbance hypothesis should be abandoned. *Trends in Ecology & Evolution*, **2013**, 28(2), 86–92. DOI:10.1016/j.tree.2012.08.014

Gómez-Ramírez, J., Ávila-Villanueva M., Ángel Fernández-Blázquez M. Selecting the most important self-assessed features for predicting conversion to mild cognitive impairment with random forest and permutation-based methods*. Scientific Reports* 10.1 **2020**, 1-15.

Górniak, A. Typologia i aktualna trofia jezior WPN. W: A. Górniak (red.), Jeziora Wigierskiego Parku Narodowego. Aktualna jakość i trofia wód. Uniwersytet w Białymstoku, Zakład Hydrobiologii. *Wydawnictwo Uniwersytetu w Białymstoku*, Białystok **2006**, 128–140.

Górniak, A. Current Climatic Conditions of Lake Regions in Poland and Impacts on Their Functioning. In *Polish River Basins and Lakes Part I*, Korzeniewska, E., Harnisz, M., Eds., Springer International Publishing: Cham, **2020**, Vol. 86, pp 1–25.
https://doi.org/10.1007/978-3-030-12123-5_1.

Hassan, G. S. Within versus Between-Lake Variability of Sedimentary Diatoms: The Role of Sampling Effort in Capturing Assemblage Composition in Environmentally Heterogeneous Shallow Lakes. *J Paleolimnol* **2018**, *60* (4), 525–541.
https://doi.org/10.1007/s10933-018-0038-8.

Heggen, M. P., Birks, H. H., Heiri, O., Grytnes, J.A., Birks, H. J. B. Are Fossil Assemblages in a Single Sediment Core from a Small Lake Representative of Total Deposition of Mite, Chironomid, and Plant Macrofossil Remains? *J Paleolimnol*, **2012**, 48 (4), 669–691. https://doi.org/10.1007/s10933-012-9637-y.

Heudre, D., C. E. Wetzel, L.Moreau, B. Van de Vijver, L. Ector. On the identity of the rare *Fragilaria subconstricta* (Fragilariaceae), with *Fragilaria* species forming ribbon–like colonies shortly reconsidered. *Plant Ecology and Evolution*, **2019**, 152: 327–339.
DOI: 10.5091/plecevo.2019.1619

Hofmann, G., M. Werum, H. Lange-Bertalot,. Diatomeen im Süßwasser-Benthos von Mitteleuropa. Bestimmungsflora Kieselalgen für die ökologische Praxis. Über, **2011**, 700, 908.

Kelly, M., Urbanic, G., Acs, E., Bennion, H., Bertrin, V., Burgess, A., Denys, L., Gottschalk, S., Kahlert, M., Karjalainen, S. M., Kennedy, B., Kosi, G., Marchetto, A., Morin, S., Picinska-Fałtynowicz, J., Poikane, S., Rosebery, J., Schoenfelder, I., Schoenfelder, J., Varbiro, G. Comparing Aspirations: Intercalibration of Ecological Status Concepts across European Lakes for Littoral Diatoms. *Hydrobiologia,* **2014**, *734* (1), 125–141.
https://doi.org/10.1007/s10750-014-1874-9.

Kelly, M., Bennion, H., Burgess, A., Ellis, J., Juggins, S., Guthrie, R., Jamieson, J., Adriaenssens, V., Yallop, M. Uncertainty in Ecological Status Assessments of Lakes and Rivers Using Diatoms. *Hydrobiologia,* **2009**, *633* (1), 5–15.
https://doi.org/10.1007/s10750-009-9872-z.

Kenkel, Norm C., Orlóci L. "Applying metric and nonmetric multidimensional scaling to ecological studies: some new results." *Ecology* 67.4, **1986**, 919-928.

Kennedy, B., N. Allott. A review of the genus Brachysira in Ireland with the description of Brachysira praegeri and Brachysira conamarae, new raphid diatoms (Bacillariophyceae) from high status waterbodies. Phytotaxa, **2017**, 326: 1-27.
DOI: 10.11646/phytotaxa.326.1.1

King, L., Clarke, G., Bennion, H., Kelly, M., Yallop, M. Recommendations for Sampling Littoral Diatoms in Lakes for Ecological Status Assessments. *J Appl Phycol* **2006**, *18* (1), 15–25. https://doi.org/10.1007/s10811-005-9009-3.

Krztoń, W., Kosiba, J. Variations in Zooplankton Functional Groups Density in Freshwater Ecosystems Exposed to Cyanobacterial Blooms. Sci. Total Environ. **2020**, 730, 139044. https://doi.org/10.1016/j.scitotenv.2020.139044

Lange-Bertalot, H., G. Hofmann, M. Werum, M. Cantonati. Freshwater benthic diatoms of Central Europe: Over 800 common species used in ecological assessment, **2017**, (Vol. 942). M. G. Kelly (Ed.). Schmitten-Oberreifenberg: Koeltz Botanical Books.

Lange-Bertalot, H., Ulrich, S. Contributions to the Taxonomy of Needle-Shaped Fragilaria and Ulnaria Species. *Lauterbornia* **2014**, *78*, 1–73.

Legendre, P. Interpreting the Replacement and Richness Difference Components of Beta Diversity: Replacement and Richness Difference Components. *Glob. Ecol. Biogeogr.* **2014**, *23* (11), 1324–1334.
https://doi.org/10.1111/geb.12207.

Legendre, P., Borcard, D., Peres-Neto, P. R. Analyzing Beta Diversity: Partitioning the Spatial Variation of Community Composition Data. *Ecological Monographs* **2005**, *75* (4), 435–450. https://doi.org/10.1890/05-0549.

Legendre, P., De Cáceres, M. Beta Diversity as the Variance of Community Data: Dissimilarity Coefficients and Partitioning. *Ecol Lett* **2013**, *16* (8), 951–963.
https://doi.org/10.1111/ele.12141.

Lengyel, E. Stress and disturbance in benthic diatom assemblages. Examples from lakes and streams and from the field and laboratory studies, **2017**, (Doctoral dissertation, Pannon Egyetem).
DOI: 10.18136/PE.2016.644

Lichstein, J. W. "Multiple regression on distance matrices: a multivariate spatial analysis tool." *Plant Ecology* 188.2, **2007**, 117-131.

MacDougall, M. J., Paterson, A. M., Winter, J. G., Jones, F. C., Knopf, L. A., Hall, R. I. Response of periphytic diatom communities to multiple stressors influencing lakes in the Muskoka River watershed, Ontario, Canada. *Freshwater Science*, **2017**, *36*(1), 77-89.
DOI: 10.1086/690144?af=R

Maloufi, S., Catherine, A., Mouillot, D., Louvard, C., Couté, A., Bernard, C., Troussellier, M. Environmental Heterogeneity among Lakes Promotes Hyper β-Diversity across Phytoplankton Communities. *Freshw Biol* **2016**, *61* (5), 633–645.
https://doi.org/10.1111/fwb.12731.

Marra, R. C., Algarte, V. M., Ludwig, T. A., Padial, A. A. Diatom diversity at multiple scales in urban reservoirs in Southern Brazil reveals the likely role of trophic state. *Limnologica*, **2018**, *70*, 49-57.
DOI: 10.1016/j.limno.2018.04.001

McNaughton, S. J., Wolf L. L. Dominance and the niche in ecological systems. Science, **1970**, 167: 131-139.

Morandín-Ahuerma, I.A. Contreras-Hernández, D. A. Ayala-Ortiz, O. Pérez-Maqueo. Socio–Ecosystemic Sustainability. Sustainability **2019,** 11:3354.
Doi:10.3390/su11123354

Nidheesh, N., Nazeer, K. A., Ameer, P. M. (2020). A Hierarchical Clustering algorithm based on Silhouette Index for cancer subtype discovery from genomic data. Neural Computing and Applications, 32(15), 11459-11476.

Nõges, P., Poikane, S., Cardoso, A. C., van de Bund, W. Water Framework Directive the Way to Water Ecosystems Sustainability in Europe. *LakeLine* **2006**, *21* (1), 36–43.

Ochieng, H., Magezi, G., Gandhi, W. P., Okot-Okumu, J., Odong, R. Epipelic diatom diversity as a bioindicator in River Aturukuku, Eastern Uganda. *Plant Ecology & Diversity*, **2022**,1-16.

Ognjanova-Rumenova, N., A.Z. Wojtal, E. Sienkiewicz, I. Botev, and T. Trichkova. Biodiversity of High Mountain Lakes in Europe with Special Regards to Rila Mountains (Bulgaria) and Tatra Mountains (Poland). Diatoms: Fundamentals and Applications **2019,** pp.335-354.
DOI: 10.1002/9781119370741.ch15

Pandey, U., Pandey, J., Singh, A. V., Mishra, A. Anthropogenic drivers shift diatom dominance-diversity relationships and transparent exopolymeric particles production in River Ganga: implication for natural cleaning of river water. *Current Science* **2017**, *(00113891)* 113.5
DOI:10.1016/0377-8398(95)00070-4

Picińska-Fałtynowicz J., Soszka, H. *Fitobentos, in: Ocena Stanu Ekologicznego Wód Zlewni Rzeki Wel. Wytyczne Do Zintegrowanej Oceny Stanu Ekologicznego Rzek i Jezior Na Potrzeby Planów Gospodarowania Wodami w Dorzeczu*, Wydawnictwo IRŚ, **2011**.

Picińska-Fałtynowicz, J., Błachuta J. Wytyczne metodyczne do przeprowadzenia oceny stanu ekologicznego jednolitych części wód rzek i jezior oraz potencjału ekologicznego sztucznych i silnie zmienionych jednolitych części wód płynących Polski na podstawie badań fitobentosu. Wydawnictwo GIOŚ, Warszawa, **2008**.

Pither, J., Aarssen L. W. The evolutionary species pool hypothesis and patterns of freshwater diatom diversity along a pH gradient. Journal of biogeography **2005**, 32: 503–513.
DOI: 10.1111/j.1365-2699.2005.01194.x

Poikane, S., Kelly, M., Cantonati, M. Benthic Algal Assessment of Ecological Status in European Lakes and Rivers: Challenges and Opportunities. *Sci. Total Environ.* **2016**, *568*, 603–613. https://doi.org/10.1016/j.scitotenv.2016.02.027.

Prygiel, J., Carpentier, P., Almeida, S., Coste, M., Druart, J.-C., Ector, L., Guillard, D., Honoré, M.-A., Iserentant, R., Ledeganck, P., Lalanne-Cassou, C., Lesniak, C., Mercier, I., Moncaut, P., Nazart, M., Nouchet, N., Peres, F., Peeters, V., Rimet, F., Rumeau, A., Sabater, S., Straub, F., Torrisi, M., Tudesque, L., Van de Vijver, B., Vidal, H., Vizinet, J., Zydek, N. Determination of the biological diatom index (IBD NF T 90–354): results of an intercomparison exercise. *J. Appl. Phycol.* **2002**, *14* (1), 27–39.
https://doi.org/10.1023/A:1015277207328.

Riato, L., Leira, M. Heterogeneity of Epiphytic Diatoms in Shallow Lakes: Implications for Lake Monitoring. *Ecol. Indic.* **2020**, *111*, 105988.
https://doi.org/10.1016/j.ecolind.2019.105988.

Rimet, F., Bouchez, A., Montuelle, B. Benthic Diatoms and Phytoplankton to Assess Nutrients in a Large Lake: Complementarity of Their Use in Lake Geneva (France–Switzerland). *Ecol. Indic.* **2015**, *53*, 231–239.
https://doi.org/10.1016/j.ecolind.2015.02.008.

Ringnér, M. "What is principal component analysis?." *Nature biotechnology* **2008**, 26.3, 303-304.

Schaumburg, J., Schranz, C., Hofmann, G., Stelzer, D., Schneider, S., Schmedtje, U. Macrophytes and phytobenthos as indicators of ecological status in German lakes—a contribution to the implementation of the Water Framework Directive. *Limnologica,* **2004**, 34(4), 302-314.
DOI: 10.1016/S0075-9511(04)80003-3

Schneider, S. C., Hjermann, D. O., Edvardsen, H. Do benthic algae provide important information over and above that provided by macrophytes and phytoplankton in lake status assessment?–Results from a case study in Norway. *Limnologica*, **2019**, *76*, 28-40.
DOI: 10.1016/j.limno.2019.02.001

Sheil, D., Burslem D. Defining and defending Connell's intermediate disturbance hypothesis: a response to Fox. *Trends in Ecology & Evolution*, **2013**, 28(10), 571–572. DOI:10.1016/j.tree.2013.07.006

Soininen, J. A Quantitative Analysis of Species Sorting across Organisms and Ecosystems. *Ecology* **2014**, *95* (12), 3284–3292.
https://doi.org/10.1890/13-2228.1.

Soininen, J., Heino, J. Variation in niche parameters along the diversity gradient of unicellular eukaryote assemblages. *Protist*, **2007**, *158*(2), 181-191.
DOI: 10.1016/j.protis.2006.11.002

Specziár, A., Árva, D., Tóth, M., Móra, A., Schmera, D., Várbíró, G., Erős, T. Environmental and Spatial Drivers of Beta Diversity Components of Chironomid Metacommunities in Contrasting Freshwater Systems. *Hydrobiologia* **2018**, *819* (1), 123–143.
https://doi.org/10.1007/s10750-018-3632-x.

Stoermer, E. F., J. P. Smol. Applications and uses of diatoms: prologue. The diatoms: Applications for the environmental and earth sciences (1st ed.), New York: Cambridge University Press, **1999**, 3–10.

Strayer, D. L., Findlay, S. E. Ecology of freshwater shore zones. *Aquatic Sciences* **2010**, 72: 127–163.
DOI: 10.1007/s00027-010-0128-9

Taylor, D. R., Aarssen, L. W., Loehle, C. On the relationship between r/K selection and environmental carrying capacity: a new habitat templet for plant life history strategies. Oikos, **1990**, 239-250.

Tibshirani R., Walther, G. Cluster validation by prediction strength. *J Comput Graph Stat*, **2005**, 14: 511–528.
DOI: 10.1198/106186005X59243

Van Dam, H., Mertens, A., Sinkeldam, J. A coded checklist and ecological indicator values of freshwater diatoms from the Netherlands. *Netherland Journal of Aquatic Ecology*, **1994**, 28: 117–133.
DOI: 10.1007/BF02334251

Van Dam, H., On the use of measures of structure and diversity in applied diatom ecology. *Nova Hedwigia*, **1982**, 73: 97–115.

Van de Vijver, B. Analysis of the Type Material of Navicula Brachysira Brébisson with the Description of Brachysira Sandrae, a New Raphid Diatom (Bacillariophyceae) from Iles Kerguelen (TAAF, Sub-Antarctica, Southern Indian Ocean). *Phytotaxa* **2014**, *184* (3), 139. https://doi.org/10.11646/phytotaxa.184.3.3.

Van Den Wollenberg, A. L. "Redundancy analysis an alternative for canonical correlation analysis." *Psychometrika* 42.2, **1977**, 207-219.

Vilmi, A., Karjalainen, S. M., Landeiro, V. L., Heino, J. Freshwater Diatoms as Environmental Indicators: Evaluating the Effects of Eutrophication Using Species Morphology and Biological Indices. *Environ Monit Assess* **2015**, *187* (5), 243.
https://doi.org/10.1007/s10661-015-4485-7.

Wiech, A. K., Marciniewicz-Mykieta, M., Toczko, B. Stan Środowiska w Polsce. Raport 2018, Główny Inspektorat Ochrony Środowiska: Warszawa, **2018**.

Witkowski, A., Bąk, M., Kierzek, A. Zmiany warunków środowiskowych w Holocenie w jeziorze Wigry w świetle analizy diatomologicznej osadów. W: Jezioro Wigry. Historia jeziora w świetle badań geologicznych i paleoekologicznych (red. J. Rutkowski, L. Krzysztofiak), **2009**, 215-226.

Witkowski, A., T. Radziejewska, B. Wawrzyniak-Wydrowska, H. Lange-Bertalot, M. Bąk, J. Gelbrecht. Living on the pH edge: diatom assemblages of low–pH lakes in western Pomerania (NW Poland). In J. Sekbach, P. Kociolek: *The Diatom World*, **2011,** 365–384. Springer, Dordrecht.

Wojtal, A. Z., L. Ector, B. Van de Vijver, E. A. Morales, S. Blanco, J. Piatek, A. Smieja. The *Achnanthidium minutissimum* complex (Bacillariophyceae) in southern *Poland. Algological Studies*, **2011**, 136: 211–238.
DOI: 10.1127/1864-1318/2011/0136-0211

Wu, K., Zhao, W., Li, M., Picazo, F., Soininen, J., Shen, J., Zhu, L., Cheng, X., Wang, J. Taxonomic Dependency of Beta Diversity Components in Benthic Communities of Bacteria, Diatoms and Chironomids along a Water-Depth Gradient. *Sci. Total Environ.* **2020**, *741*, 140462. https://doi.org/10.1016/j.scitotenv.2020.140462.

Zawisza, E., Szeroczyńska, K. The development history of Wigry Lake as shown by subfossil Cladocera. Geochronometria, **2007**, 27(1), 67-74.

Zgrundo, A., Peszek, Ł., Poradowska, A. *Podręcznik Do Monitoringu i Oceny Rzecznych Jednolitych Części Wód Powierzchniowych Na Podstawie Fitobentosu*, OceanSense Maciej Fojcik (in Polish): Gdańsk, **2018**

Zuur, A. F., Ieno, E. N., Smith G. M. Principal coordinate analysis and non-metric multidimensional scaling. *Analysing ecological data*, **2007**, 259-264.

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Limnological Characteristics and Diatom Dominants in Lakes of Northeastern Poland

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Abstract: Determination of the relationships between environmental factors and diatom assemblages is usually made for several hundred lakes spread over a large area. However, the analysis of several lakes located near Lake Wigry also gives interesting results. Lakes in Wigry National Park (Poland) with broad similarity of geological origin show clear limnological, physical, and chemical differences. Here, we report on an investigation into how these dissimilarities influence diatom assemblages. Hierarchical Cluster Analysis showed that the studied lakes can be divided into three groups: (1) disharmonic, (2) harmonious with greater human impact on the environment, and (3) harmonious with a more limited human impact. The harmonious lakes could be divided into two groups that are mainly in line with the contents of the chloride and sulfates ions taken as indicative of human impacts on the environment. Overall, the three groups had different dominance structures, as reference to the Dominance Index (DI) made clear (mean values being: (1) –70.54%, (2) –72%, and (3) –54.58%, Generalized Linear Models with the categorical independent variable (group) showed significant differences between groups (for 1–3, 2–3) p value < 0.05). Lakes impacted by anthropopressure and disharmonic ones had the strongest dominance structure. More broadly, DI differences between the groups are consistent with the Species Pool Hypothesis (SPH), while studied differences can be said to result from natural geological dissimilarities, as well as disparate anthropogenic impacts.

Keywords: lakes; disharmonic lakes; diatoms; dominance index; peat bogs; Wigry National Park; Poland

1. Introduction

Recently observed environmental changes of anthropogenic origin are associated with the steady establishment of new relationships between the environment and ecological communities. In this regard, the past state forms the basis for current trends, just as these lead to anticipated future responses. While the unprecedented threat to global biodiversity posed by human activity is clear, we have not achieved a full understanding of how this changes in response to pressure. Nevertheless, in its recent reports, the Intergovernmental Science-Policy Platform on Biodiversity and Ecosystem Services (IPBES) announced an unprecedentedly dangerous decline in biodiversity, with limits of tolerance clearly being surpassed at present [1].

Lakes are hotspots of biodiversity [2,3] as well as systems particularly sensitive to environmental change. Furthermore, as their shore-zone areas offer so many ecosystem services, lakes can be seen as among the world's most threatened environments [4] particularly in need of change monitoring. Lakes are threatened by direct (e.g., nutrient enrichment, forestry practices, and agriculture pressure) and indirect (e.g., climate change) effects [3]. Over the last several decades, interdisciplinary studies have shown the increasing impact of climate change on lakes' water chemistry, resulting in an uncommon dangerous decline in biodiversity [1,3]. Diatoms are among the indicators also used in assessing natural and human-related features of aquatic environments [5,6]. The diversity and distribution of diatoms

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(Bacillariophyta) is mainly driven by environmental conditions, and their assemblages are particularly useful bioindicators and are frequently analyzed from sediment cores to infer past conditions [7]. Their value in water quality/environmental health assessment was appreciated so relatively long ago [8]. That usefulness can be seen to lie in the rapid and precise nature of a diatom assemblage's response to changing conditions [7], with particular importance noted where tested environments are of low pH [9,10].

The first aim of the work detailed here was to compare variables relating to physical and chemical characteristics of the different waters, and to set the results against previous data for the same area [11,12]. A comparison of this kind would encapsulate long-term changes in physical and chemical properties of waters in the lakes, and—as these conditions affect the diversity of lacustrine organisms—there was value in comparing dominant diatom taxa (in terms of both numbers of species and uniformity of abundance). Thus, the second goal of the study entailed an examination of the lakes' impacts on their diatom assemblages, with results in this regard again being set against previous findings for lakes within Wigry National Park [12–15].

Here, it is the changes in water parameters, and responses in terms of diatom dominance structure, that gain presentation, which are in line with a juxtaposition of the diatom assemblages and the physical and chemical variables characterizing ten lakes located in Wigry National Park. This area makes it possible to analyze the large Wigry lake and smaller lakes with similar climate and geological origin simultaneously, as they were connected with each other in groups in the past. Covering the Wigry Lake and adjacent lakes with research gives a chance to understand the abiotic processes occurring in time in a given area. One of the characteristic features of these lakes is the differentiated human impact to their environment

2. Materials and Methods

2.1. Study Area

Located in NE Poland, Wigry National Park includes 42 lakes with a total surface area equal to 2732 ha. The whole area is under the influence of a temperate climate transitional between the maritime and the continental [16,17], although—compared with Poland's other lake districts—this one is known for its low mean annual temperature (7.2 °C) and its potentially prolonged duration of snow cover (at 118 days per year) [18]. Lakes here are of glacial and postglacial origin, and they form a landscape first generated some 12,000 years ago. The origin is in essence uniform, as all are remnants of the last glacial period, i.e., the Weichselian Glaciation [11]. Many of the lakes studied were once part of a single larger body of water termed the late-Pleistocene Pre-Wigry. Certain lakes here, such as Białe Wigierskie, Okrągłe, Krusznik, and Muliczne form a group of postglacial accumulation and separation bodies of water. Lake Suchar III is of similar genesis, relating to a process of fragmentation that generated smaller bodies of surface water. A glacial lake formed syngenetically with the retreat of the ice sheet is in turn Lake Białe Pierciańskie, while Lake Suchar Wielki is typical of the kind associated with the melting of individual blocks of ice [11].

The mainly homogeneous soils within Wigry National Park were formed from young-glacial sedimentary rocks. Most are brown or of the pararendzina type (with a high content of calcium carbonate and skeletal particles). Only a minority of soils here are podsolic or of silt and peat [19].

The largest lake studied is Lake Wigry proper (at 2163.3 ha), while all other bodies of water are very much smaller (less than 100 ha in area). Indeed, half cover only a few hectares in area (Table 1). In terms of origin, the National Park lakes are assigned to the Wigierskie group (Lakes Wigry, Białe Wigierskie, Krusznik, Okrągłe, Muliczne, Suchar Wielki, and Wygorzele), the Huciańskie group (Lake Suchar III), and the Pierciańskie group (Lakes Wądołek and Białe Pierciańskie). Almost all are connected by rivers or small streams [20], while shore-zone vegetation is very largely formed from such helophytes as common reed (*Phragmites australis* (Cav.) Trin. ex Steud), common club-rush (*Schoenoplectus lacustris* L.), and lesser bulrush (*Typha angustifolia* L.), with a limited admixture of nympheids [12]. Humic lakes are exceptions to this rule, their shore-zone vegetation formed by floating mats comprising the roots and rhizomes of vascular plants (*Scheuchzeria palustris* L., Cyperaceae, Ericaceae), as well as peat mosses and brown mosses [17]. Overall, Wigry National Park protects a wetland area with an abundance of precious and rare organisms [21].

Table 1. Limnological characteristics of lakes (modified after [11]).

Lake	Lake Area [ha]	Max. Depth [m]	Coastline Length [m]	Direct Catchment [ha]	Catchment [ha]	Typology of Lakes	Lake Group
W	2163.3	73	63920	5159.8	45293.1	flow-through	Wigierskie
K	26.7	18	2643	70.7	70.7	exorheic	Wigierskie
BW	99.9	34	5117	329.1	329.1	exorheic	Wigierskie
M	24.1	11.3	3175	191.2	191.2	exorheic	Wigierskie
OK	13.7	13	1459	28.5	906.8	flow-through	Wigierskie
BP	6.9	24	1011	50.4	50.4	endorheic	Pierciańskie
WYG	2	3	670	63.5	63.5	endorheic	Wigierskie
SW	8.44	9.6	2066	107.1	107.1	endorheic	Wigierskie
WAD	1.09	15	474	19.4	19.4	endorheic	Pierciańskie
SIII	0.44	4	320	32.2	32,2	exorheic	Huciańskie

The ten lakes selected for study were Białe Wigierskie (BW), Białe Pierciańskie (BP), Okrągłe (OK), Muliczne (M), Krusznik (K), Suchar Wielki (SW), Suchar III (SIII), Wądołek (WAD), Wygorzele (WYG), Wigry (Krzyżańska Bay (WK), and Słupiańska Bay (WS) (Table 1, Figure 1).

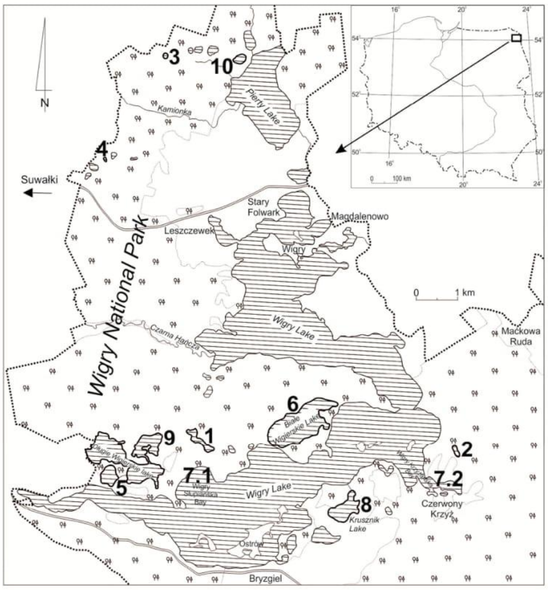

Figure 1. The study area map. Studied lakes: 1—Suchar Wielki (SW), 2—Wygorzele (WYG), 3—Wądołek (WAD), 4—Suchar III (SIII), 5—Okrągłe (OK), 6—Białe Wigierskie (BW), Wigry 7.1—Słupiańska Bay (WS), 7.2—Wigry (WK) Krzyżańska Bay, 8—Krusznik (K), 9—Muliczne (M), 10—Białe Pierciańskie (BP).

In total, 44 water samples were analyzed for their physical and chemical properties (Table 2). They were collected in spring (May 2015, May 2017), summer (September 2017), and autumn (November 2016) from 11 sampling points (one in each lake and two in the large Lake Wigry). All sampling for physical and chemical properties was done in open water, with comparable environments selected as far as possible. Conductivity and pH were measured using a YSI 6600 V2 multiparameter sonde. Water for chemical analyses was sampled 20–30 cm below the lake surface using 0.33 L polyethylene bottles and stored in the dark at 4 °C to limit ongoing chemical reactions. Ionic analyses were related to phosphate, sulfate, nitrate, fluoride, carbonate, chloride, and nitrite, as well as sodium, lithium, potassium, magnesium, ammonium, and calcium. Laboratory measurement of conductivity and pH (Table 2) involved the Dionex ion chromatograph at the laboratory of the Institute of Nature Conservation, Polish Academy of Sciences.

2.2. Statistical Data

A Hierarchical Cluster Analysis (HCA) was applied to the chemical data to identify the main sources of variation within given lakes and to produce a classification of the bodies of water analyzed. Ward's minimum variance method was used as a standard clustering procedure, while Euclidean distance served as the distance measure [22]. The Silhouette Index and Prediction Strength [23] analyzed the effectiveness of the grouping process. Principal Component Analysis was used to visualize a sample grouping in line with physical and chemical variables. Statistical analysis was performed in R 3.6.1 [24], in association with the packages: FactoMineR [25], ggplot2 [26], and factoextra [27]. The combining of HCA and PCA provided for more robust interpretation of the variability present in lake-water chemistry, as well as the change characterizing assemblages. The dominance index (after [28]) was calculated as DI = p1 + p2, i.e., the sum of the two highest abundance values (%) in a sample.

We used Generalized Linear Models (lm function) with the categorical independent variable (group) to investigate differences between groups.

2.3. Diatom Data

We sampled material from ten lakes and eleven locations (Figure 1, Tables 1 and 2). The samples were collected in autumn–spring time (November 2014, May 2015) and prepared after [29] and [30]. In total, 22 samples of periphyton (from common reed *Phragmites australis* in harmonious lakes and *Carex* spp. in disharmonic lakes) were taken. Samples were cleaned by adding 37% H_2O_2 and then heated. The reaction was completed by adding $KMnO_4$ and HCl. The cleaned diatom material dried on slides was mounted in Naphrax® synthetic resin. Then, the slides were analyzed using an Nikon Eclipse–80i microscope, with 400 diatom valves counted. Diatom identification mainly followed [5,31–38]. Dominants are defined here as the species occurring most frequently, with a relative abundance above 10%.

Table 2. Physical, chemical, and biological characteristic of the studied lakes.

Lake	Chlorides	Carbonates	Sulphates	Nitrates	Ammonium	Magnesium	Phosphates	Calcium	pH	Conductivity	Dominance Index
	[mg/L]									μS/cm	%
OK	8.45–9.99	129.30–248	28.37–30.92	0.48–1.75	0.04–0.83	11.58–15.58	0.002–0.02	46.81–73.62	7.68–8.28	338–385	68–89
M	3.52–3.85	126.70–239	21.35–24.06	0–0.37	0.02–0.22	10.14–14.45	0.002–0.011	40.21–68.25	7.52–8.26	294–342	38–59
BP	2.28–2.94	186.77–292	3.15–5.63	0–0.35	0.02–0.32	13.88–18.95	0.002–0.01	48.46–69.45	7.63–8.23	385–401	43–64
BW	2.68–3.35	74.81–137.4	6.35–6.81	0.00–0.36	0.01–0.13	4.93–6.53	0.002–0.03	23.85–36.66	7.32–8.15	164–185	59–66
K	4.24–4.52	101.49–239	12.36–22.23	0–0.35	0.036–0.23	7.66–14.45	0.002–0.01	30.45–68.25	7.51–8.26	234–342	30–47
WK	14.92–17.05	138.99–243	21.68–23.47	0–1.26	0.012–0.12	11.91–16.07	0.002–0.01	16.05–45.67	7.56–8.23	359–402	71–73
WS	14.47–16.62	135.36–237	21.92–23.57	0–1.20	0.01–0.12	11.80–16.08	0.003–0.03	16.08–43.93	7.60–8.15	350–388	61–73
WYG	1.40–2.03	8.44–35.71	0.02–1.23	0.00–0.40	0.20–1.26	0.00–0.49	0.01–0.03	0.85–2.01	3.7–6.11	20–25	89–94
WAD	1.27–1.35	17.56–18.60	0.02–0.56	0–0	0.00–0.20	0.77	0.005–0.04	2.67–3.35	4.65–5.31	20–23	63–76
SIII	0.86–8.44	9.04–18.45	0.02–1.11	0–0	0.08–0.20	0.00–0.46	0.02–0.05	0.59–1.74	3.6–6.8	22–23	79–83
SW	0.97–1.28	8.66–39.66	2.23–2.68	0.00–0.36	0.00–0.00	0.50–0.74	0.01–0.02	1.73–2.75	4.60–6.34	15–18	40–41

3. Results

3.1. Physical and Chemical Data

The physical and chemical data collected for the lakes (Table 2) showed the most marked variation in the case of water conductivity [from 15 to 402 µS/cm (in SW and WK respectively)], as well as pH [range 3.6 to 8.3 (at SIII and OK)]. Carbonates were present at the highest concentration in Lake BP (at 292 mg/L), while the lowest levels were characterized in Lake SW (8.66). The content of sulfate ions varied from 0.02 (WYG, SIII) to 30.92 mg/L (OK), chloride ions varied from 0.86 (SIII) to 17.05 mg/L (WK), nitrates varied from <0.0001 (SW, SIII, WAD, WYG) to 1.75 mg/L (OK), phosphates varied from 0.002 (M) to 0.05 mg/L (SIII), and calcium ions varied from 0.59 (at SIII) to 73.62 mg/L (at OK).

3.2. Statistical Data

The Hierarchical Cluster Analysis (HCA) grouped the lakes considered into three clusters, as (1) disharmonic, (2) harmonious with a greater human impact on the environment, and (3) harmonious with a limited human impact on the environment (Figure 2). The Silhouette Index (used to analyze the effectiveness of grouping, Figure 3 showed a relatively high score while mean the Silhouette width was 0.37. The prediction strength of these three clusters were estimated as 0.86 (PS > 0.8, strong support).

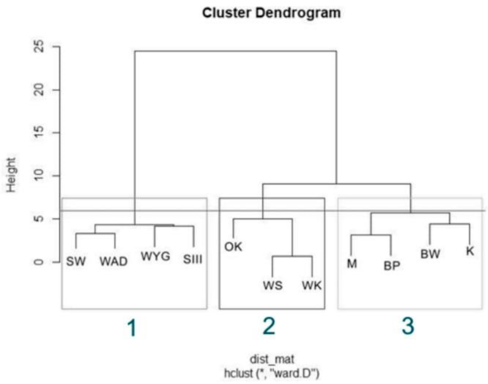

Figure 2. Hierarchical Cluster Analysis (HCA) for physical and chemical variables in the studied bodies of water, with cutting highlighting the corresponding clusters. Lakes in the first group disharmonic—box (**1**): SW, WYG, WAD, and SIII. Lakes in the second group harmonious with greater human impact on the environment—box (**2**): OK, WS, and WK. Lakes in the last group harmonious with low human impact on the environment—box (**3**) were BW, K, M, and BP.

The Principal Component Analysis (PCA) revealed two main directions of variation, with axis 1 explaining 52.4% of the total variance and axis 2 explaining an additional 12.4% (Figure 4). The strongest direction of variation (axis 1) was primarily a gradient of water conductivity, pH, and ion (mainly magnesium, carbonate, fluoride, calcium, sulfate, sodium, and chloride) concentrations (Figure 5). All of these variables correlated positively with PCA axis 1. The second gradient (of axis 2) was characterized by chloride, potassium, nitrate, lithium, phosphate, sodium, and calcium (Figure 6). PCA (Figure 4) offered a clear separation out of disharmonic lakes (group 1). None of the lakes from this group (SW, WYG, WAD, and SIII) overlapped with the members of other groups. They represent a group of distrophic lakes characterized by low pH values, limited conductivity, and low concentrations of carbonates (Table 2). In this study, group 1 showed concentrations of phosphates and ammonium that were relatively high by the standards of the ten lakes studied (Table 2).

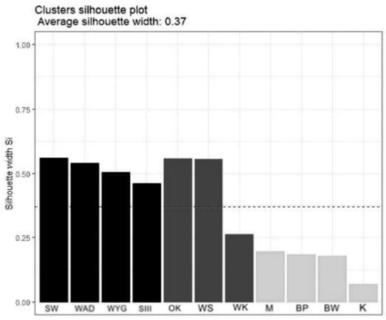

Figure 3. The Silhouette plot for HCA. Lakes in cluster 1—disharmonic lakes, lakes in cluster 2—harmonious with greater human impact on the environment, and lakes in cluster 3—harmonious with a low human impact on the environment.

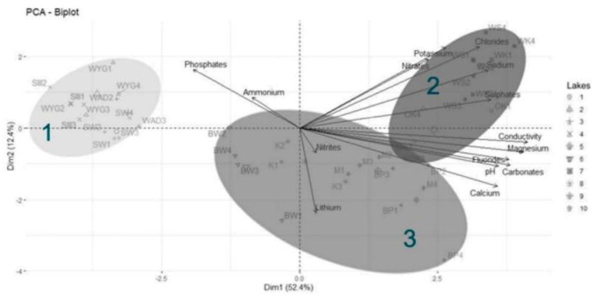

Figure 4. Principal Component Analysis (PCA) for physical and chemical variables in the studied reservoirs. Ellipses on the biplot represent cluster groups (from HCA). The cumulative explained variation (for axis 1 and 2) is 64.8%. Lakes in group 1 (disharmonic): 1—SW, 2—WYG, 3—WAD, and 4—SIII. Lakes in group 2 (harmonious with a greater human impact on the environment): 5—OK, 7—WS and WK. Lakes in group 3 (harmonious with a more limited human impact on the environment): 6—BW, 8—K, 9—M, and 10—BP. Ellipses on the biplot represent cluster groups (HCA). Water samples were collected in spring: May 2015 (notation 1 after lake abbreviation e.g., BW1), May 2017 (notation 2), summer: September 2017 (notation 3), and autumn: November 2016 (notation 4).

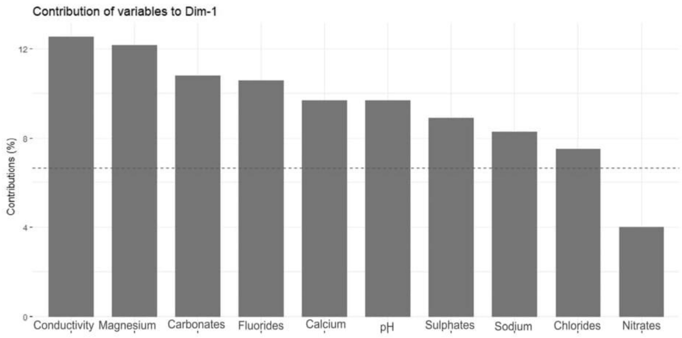

Figure 5. Contributions of variables—PCA axis 1.

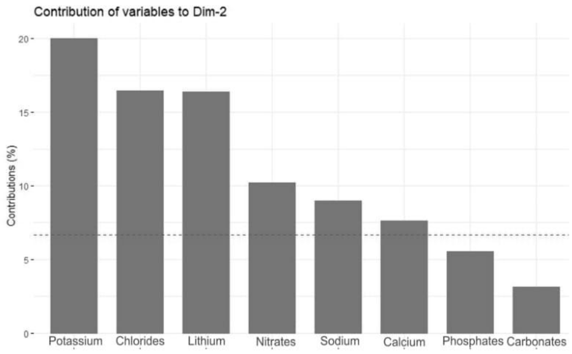

Figure 6. Contributions of variables—PCA axis 2.

The PCA and HCA both divided harmonious lakes into two groups (2 and 3), which were mainly in line with concentrations of chloride and sulfate, although nitrate content was also an important factor. Assigned to the group of harmonious lakes showing greater human impact on the environment were OK, WS and WK. These lakes were indeed characterized by their high concentrations of chloride and sulfate, but also sodium. The lakes of the third group (harmonious and with a limited human impact on the environment) were BW, K, M, and BP, and this group proved most dependent for its cohesive identity on high calcium and low orthophosphate concentrations (Figure 4). Some samples from a harmonious lake (OK2, OK3,) were in the overlapping area between groups 2 and 3, comparably so with both the HCA analysis and the Silhouette Index score.

We analyzed differences in Dominance Index between groups (Figure 7). We used Generalized Linear Models (lm function) with the categorical independent variable (group) to investigate differences between groups. It showed significant differences ($p < 0.05$) between groups 1–3 and 2–3.

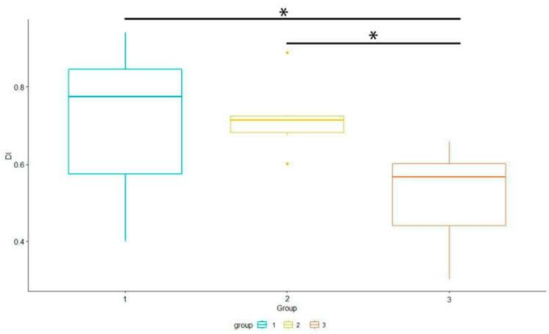

Figure 7. Differences in Dominance Index between groups. * showed significant differences ($p < 0.05$) between groups 1–3 and 2–3.

3.3. Diatom Data

Dominance within the diatom assemblages is different in the three groups (Figure 8). The most marked differences are those observed between disharmonic sites (group 1) and harmonious (groups 2 and 3). In the disharmonic group (1), the most characteristic dominant species was *Tabellaria flocculosa* (Roth) Kützing 1844. Such species as *Eunotia mucophila* (Lange-Bertalot, Nörpel-Schempp and Alles), Lange-Bertalot 2007, *E. rhomboidea* Hustedt 1950, *E. genuflexa*

Nörpel-Schempp 1996, *Stauroforma exiguiformis* (Lange-Bertalot), R.J.Flower, V.J.Jones and Round 1996, *Kobayasiella subtillissima* (Cleve) Lange-Bertalot 1999, and *Nitzschia gracilis* Hantzsch 1860 were additional dominants. The second group was solely dominated by *Achnanthidium minutissimum* (Kützing) Czarnecki 1994, *Fragilaria subconstricta* Østrup 1910, and *Encyonopsis microcephala* (Grunow) Krammer 1997. In comparison, the third group of lakes was dominated by *Achnanthidium minutissimum, Encyonopsis microcephala*, and additionally *Brachysira microcephala* (Grunow) Compère 1986, *Encyonopsis cesatii* (Rabenhorst) Krammer 1997, *Eunotia arcubus* Nörpel and Lange-Bertalot 1993, *Delicatophycus delicatulus* (Kützing) M.J.Wynne 2019, and *Staurosirella pinnata* (Ehrenberg) D.M.Williams and Round 1988. The clearest dominance structure was found in Lake WYG (DI = 93.67%) and Lake SIII (DI = 83.41%) (Figure 8). Here, the occurrence of *Tabellaria flocculosa* accounted respectively for 64.48 and 74.71% of relative abundance (Figure 8). Strong dominance in the harmonious lakes characterized WS (DI = 72.26%), and Lake OK (DI = 88.78%). Lakes K, M, and SW showed a more even structure (DI_K = 38%, DI_M = 48%, DI_{SW} = 40%).

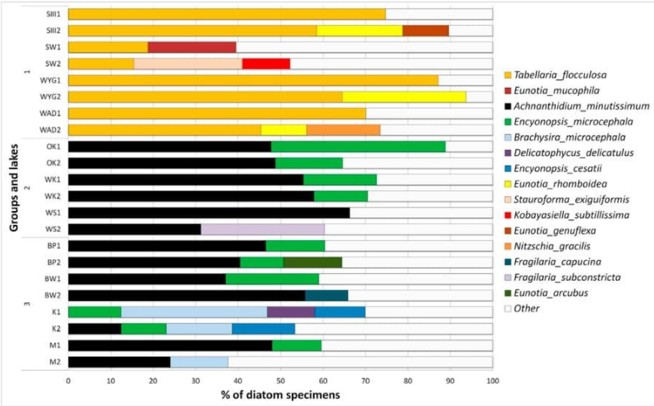

Figure 8. Relative abundance of the most numerous diatom taxa found in examined lakes group (**1**)—SW, WYG –, WAD, SIII, group (**2**)—OK, WSand WK, group (**3**)—BW, M and BP. Notation 1 (as e.g., in BW1) spring samples, notation 2 (as e.g., in BW2) autumn samples.

4. Discussion

Determination of the relationships between environments and diatom assemblages is the urgent aim of several works. Usually, these elaborations are based on a broad scale of located lakes (e.g., [39,40]). However, the kind of prediction shown in our study is clearly visible, even on a very small scale: one large lake (Wigry) and the small lakes surrounding it. Analysis of physical and chemical variables and comparison with historical data showed that the highest concentrations of both phosphate and nitrate were reached around 1996 ([12], our analysis of data). After this, a lowering of concentrations ensued, due to a change in wastewater treatment (modernization of a plant) [12]. Values for some variables, such as conductivity, have risen (Figure 9). The physical values of water and ion concentration were undoubtedly influenced by elevated temperature and very low rainfall during the research (2014–2015). Furthermore, biological variables such as the quality of macrophytes (as measured using the Ecological State Macrophyte Index—ESMI [41]) and the structure of the littoral zone (area covered by littoral vegetation) showed deterioration [12]. Similar aggravation in the quality of charophytes was reported [42] in the surrounding districts. These are indicators of human impact on the environment [41]. According to Górniak [12], the human impact in the study area was mostly attributed to a lack of separators in rainwater drainage and problems with wastewater treatment in buildings located far (up to 15km) from sewerage. Fish farming and increased tourism pressure reflecting economic growth in Poland make these processes much more complicated. Similar changes were reported by Siuda et al. [43] in the Mazurian Lake District. Siuda et al. [43] attributed the lowered concentrations of phosphate and nitrate and retreat in terms of the trophic state of lakes to economic collapse (in the 1980s–1990s), with a change in wastewater treatment and the collapse of unsustainable agriculture also involved.

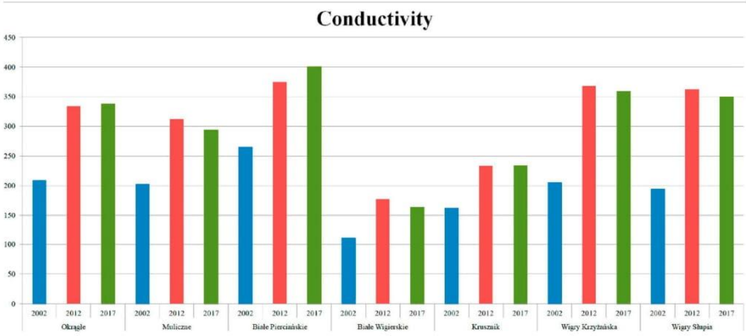

Figure 9. Changes in conductivity: our results from summer in comparison with data from 2011/2012 and 2002 (modified after Górniak (Ed.), 2014 and Górniak, 2006).

However, the reductions in concentrations of phosphate and nitrate that were noted have turned back into increases, with this attributed by Siuda et al. [43] to the problem of concentrated wastewater treatment. This is comparable to the Wigry lake case in our study area. The Siuda et al. [43] prediction is that the deposition of all treated sewage from over a larger area into bodies of water or rivers is what has been causing increased concentrations of nutrients, given that treated wastewater still has more phosphate and nitrate than naturally occurring water. Thus, the solution advocated is dispersed wastewater treatment (with water first clarified near the households that generate it, in small wastewater treatment stations). In our study area, such a solution was implemented at least in part (with wastewater treatment at the village of Bryzgiel), and the outcomes were positive. It is possible that these changes are related to the increase in the annual temperature of this area. In comparison with other lake districts in Poland, Wigry National Park continues to have lakes of good water quality [12,44,45]. The statistical analysis (PCA and HCA) in relation to water physical and chemical variables allowed the lakes under study to be assigned to three groups, i.e., (1) disharmonic, (2) harmonious with a greater human impact on the environment, and (3) harmonious with a more limited human impact on the environment. Group 1, firmly distinguished in our analysis, comprises dystrophic lakes SW, SIII, WAD, and WYG. Two of these lakes (SW and WYG) are in the Wigry group, while the others form part of the Lake Pierciańskie group (WAD) and the Lake Huciańskie group (SIII) (Table 1). Thus, while origins are different, the endorheic character is similar (except in the case of WAD). Lake SW developed in connection with melting blocks of ice, while Lake II was created during a process of fragmentation that created smaller surface bodies of water. Lake WAD came into being as the ice sheet cover of Lake BP retreated, while Lake WYG is a remnant of the Pre-Wigry lake that once existed [11,20]. Today, these lakes represent a group that is cohesive from the ecological point of view.

Water samples are characterized by low pH, low values for electrolytic conductivity, and water of a brown-yellow hue. As Górniak [11] reported, a strong shift in one variable (here a low pH value) leaves lakes unbalanced and switched toward a disharmonic path of evolution. The low pH values are in fact natural, reflecting the ombrotrophic sourcing of water. The catchments of these lakes are mainly covered by coniferous forest [12,16], so an increased supply of allochthonous matter leaves lakes rich in humic acids [11]. The range of the photic zone is reduced in consequence, with the spectrum of light penetrating the different layers of water changed, and stratification is also affected [46].

On the other hand, the chelation properties of the humic acids influence the physical and chemical variables of water, while the environment offers an additional selection factor where diatoms are concerned, as the presence of humic acids lowers concentrations of silica in water [9]. This disturbs the production of frustules, especially in early stages of development [9,47]. Only adaptable species of diatom can survive in such environments. In the disharmonic group of lakes, the most characteristic dominant species is *Tabellaria flocculosa*, which is often in fact abundant in acidic, soft waters [48]. Other species found to be abundant in the disharmonic group of lakes (group 1) were *Eunotia mucophila, E. rhomboidea, E. genuflexa, Stauroforma exiguiformis, Kobayasiella subtillissima*, and *Nitzschia gracilis*. Environments of low pH are joined by peatlands as important refugia for rare species [9,10]. Taxa from the first group reported for the first time from Wigry National Park are *Eunotia genuflexa, E. mucophila, E. rhomboidea, Kobayasiella subtillissima, Nitzschia gracilis*, and *Stauroforma exiguiformis*.

The division of the harmonious lakes into two groups was mainly a reflection of the presence of chloride and sulfate, but also nitrate. Chloride and sulfate in inland waters are usually of anthropogenic origin [12,49]. Lakes from the second group (with greater anthropopressure) are OK, WK, and WS. These lakes fall within the Wigry group and are remnants of the old, Pre-Wigry lake [11,20]. Catchments of group 3 (less affected by anthropopressure) are mostly covered in forest, with anthropopressure limited, while Lake BW (one of the group 3 lakes) enjoys special protection within Wigry National Park (being within the Strict Nature Reserve). From an origin point of view, group 3 represents the Wigierskie group (K, M, BW) and the Pierciańskie group (BP). Some samples from the harmonious lakes (groups 2 and 3) were collected in an overlapping area (OK) in PCA, which is comparable with the HCA analysis and Silhouette Index score. Analysis of biological and physical and chemical variables by Górniak [12] showed that the different indexes divide lakes from groups 2 and 3 differently. The lakes studied are subject to various kinds of anthropopressure both now and from the historical point of view. For example, Lake K has an agricultural catchment, while Lake OK has a rural catchment [12], with the result that the waters have excesses of different ions. This explains the lower Silhouette width and overlapping of these two groups—the lakes form a gradient, not two groups straightforwardly. Group (2)—harmonious with a greater human impact on the environment—was dominated by *Achnanthidium minutissimum, Fragilaria subconstricta*, and *Encyonopsis microcephala*. Only *Achnanthidium minutissimum* is observed in a wide range of environmental conditions, while *Fragilaria subconstricta* occurs in mesotrophic, slightly alkaline environments, and *Encyonopsis microcephala* is present in oligotrophic to slightly eutrophic conditions [38,46]. These taxa are more resistant to slight anthropopressure and a higher nutrient content than less common taxa from the third group. In comparison, the lakes of group (3)—harmonious with a lower human impact on the environment—were dominated by *Achnanthidium minutissimum* and *Encyonopsis microcephala*, and additionally by *Brachysira microcephala, Encyonopsis cesatii, Eunotia arcubus, Delicatophycus delicatulus*, and *Staurosirella pinnata*. These species are mostly indicative of oligotrophic to mesotrophic waters, which are rich in calcium bicarbonate [48]. This is comparable with our findings (the third group is characterized by more limited anthropopressure and lower concentrations of nitrate and phosphate).

Lake monitoring is very important, especially due to proven changes in nutrient status and the non-straightforward explanation of these processes [12]. Taxa from group 3 not reported hitherto from Wigry National Park were *Delicatophycus delicatulus, Encyonopsis cesatii, Fragilaria subconstricta*, and *Eunotia arcubus* [9,11,13–15].

The seasons were chosen in which diatoms have the highest abundance in temperate climate, due to minimizing the impact of competition on the biodiversity of diatoms. In our analysis of diatom dominance structure, we can see the clearest cases (in relation to DI) characterizing lakes WYG (DI 89–94) and SIII (DI 79–83). The marked dominance there was obviously related to lower species richness and more limited evenness. As Witkowski et al. [9] stated, when pH values fall, species richness is curtailed, and the structure of diatom assemblages can be changed profoundly. Such environments of lowered resilience are susceptible to stressors other than an excess of hydrogen ions e.g., the expansion of *Gonyostomum semen* (Ehrenberg) Diesing 1866 reported [50,51]. On the other hand, low diversity in the distrophic group can be related to low conductivity (as reported in [52]). It is worth consideration to look at the outlier in this group—Lake SW showed a more even structure (mean value—40%). Our observations and the processes of invasion by nympheids [12] may be related, but this hypothesis demands further analysis. Despite reports regarding HDI (the Hydrochemical Dystrophy Index) [11,12], some physical, chemical, and biological variables of Lake SW show differences from other dystrophic lakes, encouraging some scientists to describe it as moderately dystrophic [53]. The unique features of lake SW are also described by Karpowicz et al. [51]. Their data show a lower abundance of *Gonyostomum semen* and a higher biomass of zooplankton in Lake SW in comparison to the lower biomass in the other dystrophic lakes, as well as a deep and oxygenated epilimnion in contrast to lower oxygen concentration in other disharmonic lakes. The changes in diatom dominance structure are not linked solely with pH value; the pH values noted for Lake SW are not the highest in the dystrophic group ([11,12] and our data), yet the structure and low DI repeats in other seasons (average pH by the standards of the dystrophic group yet the most limited dominance). Another influencing factor is presumably present.

Generalized Linear Models showed significant ($p < 0.05$) differences in Dominance Index in two groups of harmonious lakes. Strong dominance (DI > 70%) in the context of harmonious lakes was found for Lake WS and WK, and for Lake OK. On the other hand, Lakes K and M (M) show a more even distribution of dominants ($DI_K < 50$, $DI_M < 60$). As Van Dam [54] reports, such a peak of diversity can be connected with an intermediate level of disturbance. The expected structure of diatom assemblages of natural lakes is one or two strongly dominant taxa, a few subdominants, and rare species in the minority. For example, as a disturbance, a temporal drought [55] is introduced; then, the dominance structure becomes more even—with a few dominants and more rare species. This can be explained by reference to a change from one environment with its specialists to another with different specialists. Thus, peak species richness is connected with a rebuilding of the structure of the diatom assemblage. More intensive disturbance in turn renders the environment more homogeneous and thus generates a collapse in species richness. Such effects are in line with the Intermediate Disturbance Hypothesis [56], although this structure of dominance repeats between seasons, raising a question as to whether these are repeated disturbances (consistent rebuilding), stressors, or other factors. In our case, Lake BP and Lake SW had an even structure (lower DI), yet they are especially protected or hard to reach in the field. The IDH has been a subject of heated debate in recent years (e.g., [36,57,58]). Anthropogenic disturbances (as in group 2) are known to reduce biodiversity [59] and are shifting in species composition toward taxa thriving in nutrient-rich conditions ([60]—for nutrient enrichment). In our case, changes in the diatom dominance structure can be connected with the changing physical and chemical properties of water in lakes to a more evolutionary common one (e.g., from electrolyte-poor and acidic to circumneutral water, as between groups 1 and 3). In such environments, there are more specialists available due to an evolutionarily longer time for adaptation [61,62]. This point of view is consistent with the Species Pool Hypothesis [61,62]. In groups, we can see similar patterns, as the third group—of intermediate status (in terms of pH and nutrient content) and with lower anthropopressure—shows a depression in the Dominance Index. Moreover, as research concerning our study area showed, high pH and a relatively high mineralization or oligotrophy were more common in this area [63,64]. Recent research reports diatoms behavior as consistent with SPH [65]; however, more evidence will be more conclusive. Samples with a high Dominance Index were inhabited by either generalists or specialists (group 1 and 2), which is consistent with observations reported by Soininen and Heino [66]; however, samples in group 3, which were more diverse, were dominated firstly by generalists and then specialists (contradictory to Soininen and Heino [66], but this was consistent with other analyses on lakes, as in e.g, [67,68]).

Strong pressure exerted by tourism and fish farming and problems with sewage treatment plants all potentially pose dangerous threats to this region. Moreover, the lakes are threatened by indirect effects (climate change). Therefore, further monitoring of physical, chemical, and biological changes here is a necessity.

5. Conclusions

The lakes in Poland's Wigry National Park are diversified in size, depth, and trophic status. The lakes belong to the Wigierskie, Huciańskie, and Pierciańskie groups and show broad heterogeneity in chemical variables and diatom dominance structure. The variability reflects differences in both the geological and anthropological history of the studied lakes. Hierarchical Cluster Analysis (HCA) and Principal Component Analysis (PCA) clearly separated off disharmonic lakes (of low pH and rich in allochthonous matter), as opposed to harmonious ones. The latter were divided into groups indicating human impact on the environment as either high or low, with this assessment depending mainly on content of chloride and sulfate ions. The comparison of dominant diatom taxa is justified, as physical and chemical variables describing water are important factors affecting numbers of species and the uniformity of abundance among organisms that inhabit them. In the disharmonic group of lakes, the most distinctive dominant species was *Tabellaria flocculosa*, with harmonious lakes in turn being dominated by *Achnanthidium minutissimum* and *Encyonopsis microcephala*. The first group was dominated by acidophilous diatoms, the second group was dominated by species tolerating a wide spectrum of habitat conditions, and the third group was dominated by more sensitive kinds that prefer oligotrophic to mesotrophic waters. Lakes impacted by anthropopression and disharmonic had the strongest dominance structure. The three groups had different dominance structures, given the abundance of dominants as well as the Dominant Index (mean values being: (1)—70.54%, (2)—72%, and (3)—54.58%, Generalized Linear Models with the categorical independent variable (for 1–3, 2–3) p value < 0.05). The high figure in the first case can be considered due to the more severe environmental conditions experienced (low pH and the presence of humic acids); on the other hand, the high DI in group 2 can be associated with anthropopression indicators. The differences in environmental variables and observed diatom dominance structure arise out of natural geological differences and levels of anthropogenic impact.

Author Contributions: Conceptualization, methodology, software, validation, formal analysis, investigation, M.E.-K.; writing—original draft preparation, M.E.-K. and A.Z.W.; data curation, A.Z.W. All authors have read and agreed to the published version of the manuscript.

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References

1. Morandín-Ahuerma, I.; Contreras-Hernández, A.; Ayala-Ortiz, D.A.; Pérez-Maqueo, O. Socio—Ecosystemic sustainability. *Sustainability* **2019**, *11*, 3354. [CrossRef]
2. Krztoń, W.; Kosiba, J.; Pociecha, A. The effect of cyanobacterial blooms on bio-and functional diversity of zooplankton communities. *Biodivers. Conserv.* **2019**, *28*, 1815–1835. [CrossRef]
3. Ognjanova-Rumenova, N.; Wojtal, A.Z.; Sienkiewicz, E.; Botev, I.; Trichkova, T. Biodiversity of high mountain lakes in europe with special regards to Rila Mountains (Bulgaria) and Tatra Mountains (Poland). *Diatoms Fundam. Appl.* **2019**, 335–354. [CrossRef]
4. Strayer, D.L.; Findlay, S.E. Ecology of freshwater shore zones. *Aquat. Sci.* **2010**, *72*, 127–163. [CrossRef]

5. Hofmann, G.; Werum, M.; Lange-Bertalot, H. *Diatomeen im Süßwasser-Benthos von Mitteleuropa. Bestimmungsflora Kieselalgen für die Ökologische Praxis; Über, 700 der Häufigsten Arten und Ihrer Ökologie*; Gantner: Nüziders, Austria, 2011; pp. 1–942.
6. Marra, R.C.; Algarte, V.M.; Ludwig, T.A.; Padial, A.A. Diatom diversity at multiple scales in urban reservoirs in Southern Brazil reveals the likely role of trophic state. *Limnologica* **2018**, *70*, 49–57. [CrossRef]
7. Stoermer, E.F.; Smol, J.P. Applications and uses of diatoms: Prologue. In *The Diatoms: Applications for the Environmental and Earth Sciences*, 1st ed.; Cambridge University Press: New York, NY, USA, 1999; pp. 3–10.
8. Van Dam, H. On the use of measures of structure and diversity in applied diatom ecology. *Nova Hedwig.* **1982**, *73*, 97–115.
9. Witkowski, A.; Radziejewska, T.; Wawrzyniak-Wydrowska, B.; Lange-Bertalot, H.; Bąk, M.; Gelbrecht, J. Living on the pH edge: Diatom assemblages of low–pH lakes in western Pomerania (NW Poland). In *The Diatom World*; Sekbach, J., Kociolek, P., Eds.; Springer: Dordrecht, The Netherlands, 2011; pp. 365–384.
10. Rybak, M.; Poradowska, A.; Kochman-Kedziora, N.; Peszek, Ł.; Noga, T.; Stanek-Tarkowska, J. Okrzemki (bacillariophyta) torfowiska wołosate (bieszczadzki park narodowy). *Rocz. Bieszcz.* **2018**, *26*, 169–183.
11. Górniak, A. *Typologia i Aktualna Trofia Jezior WPN*; Wydawnictwa–Uniwersytet w Białymstoku. Zakład Hydrobiology: Białystok, Poland, 2006; pp. 128–140.
12. Górniak, A. Plan Ochrony dla Wigierskiego Parku Narodowego i Obszaru Natura 2000 Ostoja PLH200004. Available online: http://www.wigry.org.pl/inf_i_rozw/plany/materialy.htm (accessed on 4 August 2019).
13. Tomaszewicz, G.H. Flora glonow wigierskiego parku narodowego. *Fragm. Florist. Geobot. Pol.* **1996**, *3*, 239–259.
14. Rakowska, B. Preliminary elaboration of the diatom communities occuring in the benthos of the Kamionka River and Lake Czarne at the Pojezierze Suwalskie (1993–1994). *Fragm. Florist. Geobot. Pol.* **1996**, *3*, 221–238.
15. Bobrukiewicz, K. Bory Tucholskie i Inne Obszary Leśne. Ochrona, Monitoring, Edukacja. Available online: https://books.google.cn/books/about/Bory_Tucholskie_i_inne_obszary_le%C5%9Bne.html?id=ejZ0PgAACAAJ&redir_esc=y (accessed on 12 July 2019).
16. Andrzejczyk, T.; Brzeziecki, B. The structure and dynamics of old-growth Pinus sylvestris (L.) stands in the Wigry National Park, north-eastern Poland. *Vegetatio* **1995**, *117*, 81–94. [CrossRef]
17. Drzymulska, D.; Zieliński, P. Phases and interruptions in postglacial development of humic lake margin (Lake Suchar Wielki, NE Poland). *Limnol. Rev.* **2014**, *14*, 13–20. [CrossRef]
18. Górniak, A. *Current Climatic Conditions of Lake Regions in Poland and Impacts on Their Functioning*; Polish River Basins and Lakes–Part. I, 1–25; Springer: Cham, Switzerland, 2020. [CrossRef]
19. Skrajna, T.; Kubicka, H. Segetal flora of the wigry national park. *Acta Agrobot.* **2011**, *64*, 119–132. [CrossRef]
20. Kostrowicki, A.S.; Baumgart-Kotarba, M. *Studium Geoekologiczne Rejonu Jezior Wigierskich*; Zakład Narodowy im. Ossolińskich: Warszawa, Poland, 1988.
21. Rutkowski, J.; Krzysztofiak, L. *Jezioro Wigry: Historia Jeziora w Świetle Badań Geologicznych i Paleoekologicznych*; Stowarzyszenie Człowiek i Przyroda: Podlaskie, Poland, 2009.
22. Boggero, A.; Zaupa, S.; Musazzi, S.; Rogora, M.; Dumnicka, E.; Lami, A. Environmental factors as drivers for macroinvertebrate and diatom diversity in Alpine lakes: New insights from the Stelvio National Park (Italy). *J. Limnol.* **2019**, *78*, 147–162. [CrossRef]
23. Tibshirani, R.; Walther, G. Cluster validation by prediction strength. *J. Comput. Graph. Stat.* **2005**, *14*, 511–528. [CrossRef]
24. R Core Team. R: A Language and Environment for Statistical Computing. R Foundation for Statistical Computing, Vienna, Austria 2013. Available online: http://www.R-project.org (accessed on 2 March 2020).
25. Husson, F.; Josse, J.; Le, S.; Mazet, J.; Husson, M.F. Package 'FactoMineR'. *Package R FactorMine* **2019**, *96*, 698.
26. Wickham, H. *Ggplot2: Elegant Graphics for Data Analysis*; Springer: Berlin/Heidelberg, Germany, 2016. [CrossRef]
27. Kassambara, A.; Mundt, F. Factoextra: Extract and visualize the results of multivariate data analyses. *R Package Version* **2017**, *1*, 337–354.
28. McNaughton, S.J.; Wolf, L.L. Dominance and the niche in ecological systems. *Science* **1970**, *167*, 131–139. [CrossRef]
29. Picińska-Fałtynowicz, J.; Błachuta, J. *Wytyczne Metodyczne do Przeprowadzenia Oceny Stanu Ekologicznego Jednolitych Części Wód Rzek i Jezior Oraz Potencjału Ekologicznego Sztucznych i Silnie Zmienionych Jednolitych Części Wód Płynących Polski na Podstawie Badań Fitobentosu*; Wydawnictwo GIOŚ Warszawa: Warsaw, Poland, 2008; pp. 1–79.

30. Zgrundo, A.; Peszek, Ł.; Poradowska, A. *Podręcznik do Monitoringu i Oceny Rzecznych Jednolitych Części Wód Powierzchniowych na Podstawie Fitobentosu*; Ocean Sense: Gdańsk, Poland, 2018.
31. Kulikovskiy, M.S.; Lange-Bertalot, H.; Witkowski, A.; Dorofeyuk, N.I.; Genkal, S.I. Diatom assemblages from Sphagnum bogs of the world. *Bibl. Diatomol.* **2010**, *55*, 1–326.
32. Wojtal, A.Z.; Ector, L.; Van de Vijver, B.; Morales, E.A.; Blanco, S.; Piatek, J.; Smieja, A. The Achnanthidium minutissimum complex (Bacillariophyceae) in southern Poland. *Algol. Stud.* **2011**, *136*, 211–238. [CrossRef]
33. Lange-Bertalot, H.; Ulrich, S. Contributions to the taxonomy of needle-shaped Fragilaria and Ulnaria species. *Lauterbornia* **2014**, *78*, 1–73.
34. Van de Vijver, B. Analysis of the type material of Navicula brachysira Brébisson with the description of Brachysira sandrae, a new raphid diatom (Bacillariophyceae) from Iles Kerguelen (TAAF, sub-Antarctica, southern Indian Ocean). *Phytotaxa* **2014**, *184*, 139–147. [CrossRef]
35. Delgado, C.; Novais, M.H.; Blanco, S.; Almeida, S.F. Examination and comparison of Fragilaria candidagilae sp. nov. with type material of Fragilaria recapitellata, F. capucina, F. perminuta, F. intermedia and F. neointermedia (Fragilariales, Bacillariophyceae). *Phytotaxa* **2015**, *231*, 1–18. [CrossRef]
36. Lengyel, E. Stress and Disturbance in Benthic Diatom Assemblages. Examples from Lakes and Streams and from the Field and Laboratory Studies. Ph.D. Thesis, University of Pannonia, Veszprém, Hungary, 2016. [CrossRef]
37. Kennedy, B.; Allott, N. A review of the genus Brachysira in Ireland with the description of Brachysira praegeri and Brachysira conamarae, new raphid diatoms (Bacillariophyceae) from high status waterbodies. *Phytotaxa* **2017**, *326*, 1–27. [CrossRef]
38. Heudre, D.; Wetzel, C.E.; Moreau, L.; Van de Vijver, B.; Ector, L. On the identity of the rare Fragilaria subconstricta (Fragilariaceae), with Fragilaria species forming ribbon–like colonies shortly reconsidered. *Plant Ecol. Evol.* **2019**, *152*, 327–339. [CrossRef]
39. Bennett, J.R.; Cumming, B.F.; Ginn, B.K.; Smol, J.P. Broad-scale environmental response and niche conservatism in lacustrine diatom communities. *Glob. Ecol. Biogeogr.* **2010**, *19*, 724–732. [CrossRef]
40. Schaumburg, J.; Schranz, C.; Hofmann, G.; Stelzer, D.; Schneider, S.; Schmedtje, U. Macrophytes and phytobenthos as indicators of ecological status in German lakes—A contribution to the implementation of the Water Framework Directive. *Limnologica* **2004**, *34*, 302–314. [CrossRef]
41. Ciecierska, H.; Kolada, A. ESMI: A macrophyte index for assessing the ecological status of lakes. *Environ. Monit. Assess.* **2014**, *186*, 5501–5517. [CrossRef]
42. Kłosowski, S.; Tomaszewicz, G.H.; Tomaszewicz, H. The expansion and decline of charophyte communities in lakes within the Sejny Lake District (north-eastern Poland) and changes in water chemistry. *Limnologica* **2006**, *36*, 234–240. [CrossRef]
43. Siuda, W.; Kaliński, T.; Kauppinen, E.; Chróst, R.J. Eutrofizacja południowej części kompleksu wielkich jezior mazurskich w latach 1977–2011. *Technol. Wody* **2014**, *3*, 48–62.
44. Bok, G.; Boguszewska, A.; Barańska, A.; Panasewicz, A.; Wiśniewska, M. Raport o Stanie Środowiska Województwa Podlaskiego 2015. Available online: http://www.wios.bialystok.pl/pdf/wios_raport2015_wersja_WWW.pdf (accessed on 6 June 2020).
45. Panek, P.; Biliński, G.; Brodowska, M.; Chełstowska, A.; Czajka, J.; Dudek, M.; Fornal, B.; Kamińska, M.; Kasprowicz, H.; Kociuba, M.; et al. Stan Środowiska w Polsce. Raport 2018. Available online: http://www.gios.gov.pl/pl/aktualnosci/521-stan-srodowiska-w-polsce-raport-2018 (accessed on 12 May 2020).
46. Górniak, A.; Jekatierynczuk-Rudczyk, E.; Dobrzyń, P. Hydrochemistry of three dystrophic lakes in Northeastern Poland. *Acta Hydrochim. Hydrobiol.* **1999**, *27*, 12–18. [CrossRef]
47. Hervé, V.; Derr, J.; Douady, S.; Quinet, M.; Moisan, L.; Lopez, P.J. Multiparametric analyses reveal the pH-dependence of silicon biomineralization in diatoms. *PLoS ONE* **2012**, *7*, e46722. [CrossRef] [PubMed]
48. Lange-Bertalot, H.; Hofmann, G.; Werum, M.; Cantonati, M. *Freshwater Benthic Diatoms of Central Europe: Over 800 Common Species Used in Ecological Assessment*; Koeltz Botanical Books: Hochtaunuskreis, Germany, 2017.
49. Bennion, H.; Sayer, C.D.; Tibby, J.; Carrick, H.J. Diatoms as indicatorsof environmental change in shallow lakes. In *The Diatoms: Applications for the Environmental and Earth Sciences*; Cambridge University Press: Cambridge, UK, 2010. [CrossRef]
50. Pęczuła, W.; Grabowska, M.; Zieliński, P.; Karpowicz, M.; Danilczyk, M. Vertical distribution of expansive, bloom-forming algae Gonyostomum semen vs. plankton community and water chemistry in four small humic lakes. *Knowl. Manag. Aquat. Ecosyst.* **2018**, *419*, 28. [CrossRef]

51. Karpowicz, M.; Zieliński, P.; Grabowska, M.; Ejsmont-Karabin, J.; Kozłowska, J.; Feniova, I. Effect of eutrophication and humification on nutrient cycles and transfer efficiency of matter in freshwater food webs. *Hydrobiologia* **2020**. [CrossRef]
52. Pestryakova, L.A.; Herzschuh, U.; Gorodnichev, R.; Wetterich, S. The sensitivity of diatom taxa from Yakutian lakes (north-eastern Siberia) to electrical conductivity and other environmental variables. *Polar Res.* **2018**, *37*, 1485625. [CrossRef]
53. Szczęsny, B.; Majecki, J. Variations in the composition of Trichoptera communities associated with lakes of different trophy in the Wigry National Park (NE-Poland). *Nova Suppl. Entomol.* **2002**, *15*, 581–593.
54. Van Dam, H.; Mertens, A.; Sinkeldam, J. A coded checklist and ecological indicator values of freshwater diatoms from The Netherlands. *Neth. J. Aquat. Ecol.* **1994**, *28*, 117–133. [CrossRef]
55. Calapez, A.R.; Elias, C.L.; Almeida, S.F.; Feio, M.J. Extreme drought effects and recovery patterns in the benthic communities of temperate streams. *Limnetica* **2014**, *33*, 281–296. [CrossRef]
56. Connell, J.H. Diversity in tropical rain forests and coral reefs. *Science* **1978**, *199*, 1302–1310. [CrossRef]
57. Fox, J.W. The intermediate disturbance hypothesis should be abandoned. *Trends Ecol. Evol.* **2013**, *28*, 86–92. [CrossRef]
58. Sheil, D.D.; Burslem, F.R.P. Defining and defending Connell's intermediate disturbance hypothesis: A response to Fox. *Trends Ecol. Evol.* **2013**, *28*, 571–572. [CrossRef]
59. Pandey, U. Anthropogenic drivers shift diatom dominance-diversity relationships and transparent exopolymeric particles production in River Ganga: Implication for natural cleaning of river water. *Curr. Sci.* **2017**, *00113891*, 113. [CrossRef]
60. Schneider, S.C.; Hjermann, D.O.; Edvardsen, H. Do benthic algae provide important information over and above that provided by macrophytes and phytoplankton in lake status assessment?–Results from a case study in Norway. *Limnologica* **2019**, *76*, 28–40. [CrossRef]
61. Taylor, D.R.; Aarssen, L.W.; Loehle, C. On the relationship between r/K selection and environmental carrying capacity: A new habitat templet for plant life history strategies. *Oikos* **1990**, *58*, 239–250. [CrossRef]
62. Pither, J.; Aarssen, L.W. The evolutionary species pool hypothesis and patterns of freshwater diatom diversity along a pH gradient. *J. Biogeogr.* **2005**, *32*, 503–513. [CrossRef]
63. Witkowski, A.; Bąk, M.; Kierzek, A. Zmiany warunków środowiskowych w Holocenie w jeziorze Wigry w świetle analizy diatomologicznej osadów. In *Jezioro Wigry Historia Jeziora w Świetle Badań Geologicznych i Paleoekologicznych*; Rutkowski, J., Krzysztofiak, L., Eds.; Stowarzyszenie "Człowiek i Przyroda": Podlaskie, Poland, 2009; pp. 215–226.
64. Zawisza, E.; Szeroczyńska, K. The development history of Wigry Lake as shown by subfossil Cladocera. *Geochronometria* **2007**, *27*, 67–74. [CrossRef]
65. González-Trujillo, J.D.; Pedraza-Garzón, E.; Donato-Rondon, J.C.; Sabater, S. Ecoregional characteristics drive the distribution patterns of neotropical stream diatoms. *J. Phycol.* **2020**. [CrossRef]
66. Soininen, J.; Heino, J. Variation in niche parameters along the diversity gradient of unicellular eukaryote assemblages. *Protist* **2007**, *158*, 181–191. [CrossRef]
67. Chen, W.; Wang, Q.; Xia, S.; Pei, G. Comparative responses of dominant and subordinate periphytic diatom species to environmental gradients in Donghu Lake, China. *Int. Rev. Hydrobiol.* **2016**, *101*, 182–192. [CrossRef]
68. MacDougall, M.J.; Paterson, A.M.; Winter, J.G.; Jones, F.C.; Knopf, L.A.; Hall, R.I. Response of periphytic diatom communities to multiple stressors influencing lakes in the Muskoka River watershed, Ontario, Canada. *Freshw. Sci.* **2017**, *36*, 77–89. [CrossRef]

ARTYKUŁ 2:

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Influence of Selected Environmental Factors on Diatom β Diversity (Bacillariophyta) and the Value of Diatom Indices and Sampling Issues

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Abstract: Human impacts and environmental climate changes have led to a progressive decline in the diversity of diatoms in lakes in the recent past. The components of β diversity (e.g., species turnover and nestedness) and underlying factors are still poorly understood. Here, we report an investigation of two alternative approaches—beta diversity (β diversity) partitioning and local contribution to β diversity (LCBD)—including their responses to selected environmental factors and representativeness of samples in estimating the ecological fitness of a lake. The β diversity of diatoms and their local contributions could be explained by the effects of environmental variables ($p < 0.01$). The random forest method showed the most contribution to the variance for NO_3^-, Cl^-, and SO_4^{2-}. PERMANOVA as well as a network analysis in JASP (Jeffrey's Amazing Statistics Program) showed significant differences between the seasons in diatom assemblages and in the diatom index for Polish lakes (IOJ). Our findings provide insights into the mechanisms responsible for community organizations along environmental gradients from the perspective of β diversity components, and mechanisms of the indication value of diatoms for lakes; the results could be used especially by countries implementing ecological assessments.

Keywords: β diversity; turnover; nestedness; lakes; diatoms; diatom index for Polish lakes; Wigry National Park; Poland

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1. Introduction

The response of organisms to habitat loss is one of the most important directions of modern research. The destruction of the natural habitats of lakes is associated with a deterioration in water quality, reflecting a decline in the value of β diversity. Poor water quality is reflected in the values of diatom indices. The low quality of habitats is a threat to many aquatic species, although some can survive a degree of degradation. Understanding the mechanisms responsible for community organization along environmental gradients from the perspective of β diversity components is now a central issue in ecology and conservation biology [1]. β diversity, its components (e.g., species turnover and nestedness), and the underlying drivers remain poorly understood, even though β diversity plays an important role in explaining ecological processes, especially for cross-scale biodiversity patterns [2]. Studies of metacommunity processes as well as of β diversity and its components are gaining more and more attention [3]. An estimation of β diversity allows us to measure the differences between the communities present in every place, considering the identities of all the species [4]. It gives a unique opportunity for understanding different layers of environmental conditions and water quality. The β diversity proposed by the Balsega [5] framework is based on a presence-absence matrix and can be divided into the components of turnover (species replacement—one species replaces another with no change in species richness) and nestedness (diversity differences due to species gain or loss). A different approach framework proposed for analyzing β diversity, especially at the local scale, was developed by Legendre and De Cáceres [6] for matrices with a quantified species contribution (LCBD—local contribution to β diversity). The LCBD index can also be partitioned into replacement (equivalent to turnover) and nestedness. Species turnover/replacement is impacted by environmental filtering, competition, and historical events [7], while species nestedness is impacted by species thinning and other ecological processes (human impact, physical barriers, etc.) [8]. All these methods are useful in the analysis of habitat changes and environmental conservation.

The Water Framework Directive requires classifying all surface waterbodies according to their ecological status, and it gives us a more holistic view [9]. The state of waterbodies is examined so that a tipping point before serious deterioration can be caught and waterbody management can be thus implied [10]. Diatoms are considered to be among the best groups of biota used for waterbody assessments [11–14]. Diatom-based metrics show strong responses to nutrient gradients [15–17]. However, many countries, including Poland, still have problems with the successful implementation of diatom indices for lakes [18–23], and many others are also about to implement ecological indicators to their environmental inspections. Custom river indexes are used for many lake water assessments [18,20]. The diatom index for Polish lakes (IOJ) rarely specifies assessments other than very good or good; even less frequent are situations when the IOJ is responsible for the final evaluation [21–23]. Other biological elements show a more varied distribution [21–23]. Sources of imperfections in the diatom index of Polish lakes [24] were revised by Zgrudno et al. [25], making the method easier for users; however, many problems remain unresolved. For example, only one sample from a lake is required to be collected in one year in Poland. One sample per lake is consistent with the recommendations made by the authors of [26]—one sampling site distant from source of pollution is thought to be sufficient for the needs of water management. Many countries are testing phytobenthos more than once a year and using more than one sample per lake [18,27,28], so we hypothesized that one-time sampling from a lake may be a source of error in the field method. In support, some studies have shown differences in diatom assemblages within lake surveys [29–33]. The variability of phytobenthos communities is different for the studied lakes and is additionally related to the seasons of the year [31,32]. This variability is not always related to the availability of nutrients. The hydrological regime, light, temperature, and grazing practices also affect phytobenthos [10,34].

The aim of our research was to investigate the influence of the environmental factors of lakes on the β diversity of diatoms and the diatom index, and to assess the representativeness of one lake sample per year.

2. Materials and Methods

2.1. Study Area

This study took place in Wigry National Park, in Northeast Poland, and included four lakes—Białe Wigierskie (BW), Krusznik (K), Okrągłe (O) and Wigry (W) (Figure 1, Table 1). The study area is under the influence of a temperate climate, transitional between the maritime and the continental [35,36]. The studied lakes are of glacial and postglacial origin and are remnants of the Weichselian glaciation [37]. The lakes differed in their limnological, physical, and chemical features (Tables 1 and 2); however, they were all characterized by harmonic evolution. The biggest lake was Wigry—at 2163.3 ha—and the smallest was Okrągłe lake—at 13.7 ha. The direct catchment of these lakes also differed, despite the fact that they are all in National Park. Okrągłe and Wigry were more affected by human impact; on the other hand, Krusznik was impacted by extensive agriculture. The direct catchment of Białe Wigierskie lake was under strict protection. These differences gave us the opportunity to test the β diversity as well as the diatom index for Polish lakes (IOJ) for a relatively wide spectrum of variables, despite the small study area.

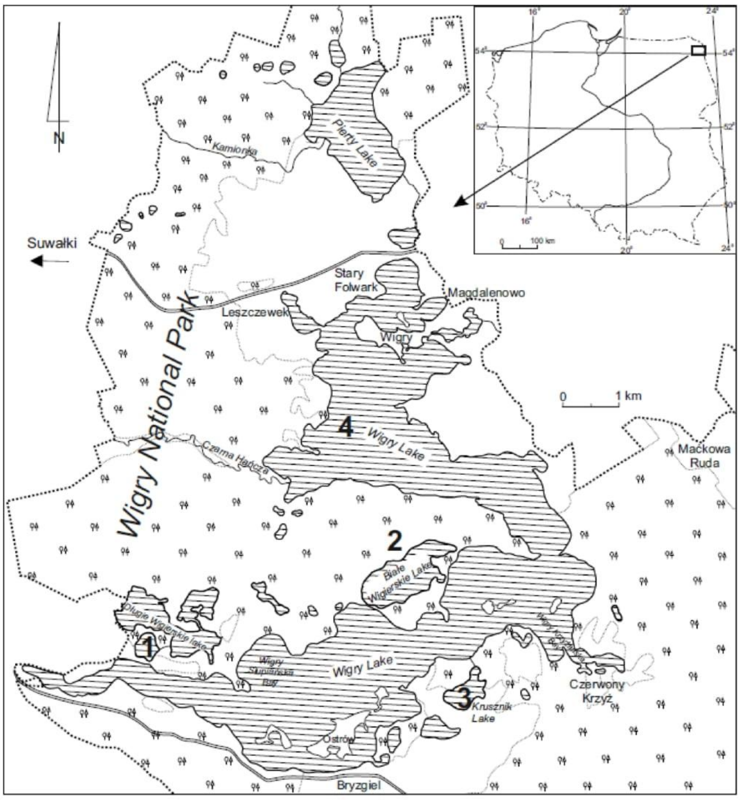

Figure 1. Map of the study area; 1—Okrągłe lake (O), 2—Białe Wigierskie lake (BW), 3—Krusznik lake (K), and 4—Wigry lake (W).

Table 1. Limnological characteristics of studied lakes.

Name of Lake	Lake Area (ha)	Max. Depth (m)	Coastline Length (m)	Direct Catchment (ha)	Catchment (ha)
1. Okrągłe (O)	13.7	13	1459	28.5	906.8
2. Białe Wigierskie (BW)	99.9	34	5117	329.1	329.1
3. Krusznik (K)	26.7	18	2643	70.7	70.7
4. Wigry (W)	2163.3	73	63,920	5159.8	45,293.1

Table 2. Physical and chemical characteristics of studied lakes. O—Okrągłe lake; BW—Białe Wigierskie lake; K—Krusznik lake; WK—Wigry lake Krzyżańska bay; WS—Wigry lake Słupiańska bay; WP—Wigry lake Piaski; and WB—Wigry lake Bryzgiel.

Lakes	Cl^-	CO_3^{2-}	SO_4^{2-}	NO_3^-	NH_4^+	Mg2$^+$	PO_4^{3-}	Ca^{2+}	pH	Conductivity
	(mg/L)									(µS/cm)
O	4.10–9.96	103.52–144.61	12.21–28.56	0.03–0.22	0.036–0.23	9.17–16.17	0.000–0.003	34.85–73.46	7.31–8.26	231–361
BW	2.70–2.78	73.39–94.93	5.92–6.34	0.00–0.02	0.01–0.13	5.53–6.96	0.000–0.005	25.05–38.07	7.28–8.44	161–174
K	4.46–10.21	127.44–129.29	12.33–27.23	0.22–0.24	0.036–0.23	8.63–13.85	0.002–0.001	35.87–57.07	7.29–8.33	251–327
WK	16.31–17.06	129.32–147.83	21.68–23.47	0.01–0.09	0.012–0.12	13.09–16.07	0.000–0.005	42.05–67.50	7.55–8.25	349–374
WS	15.82–16.50	119.93–155.12	20.61–22.39	0.01–0.05	0.01–0.12	12.96–17.14	0.000–0.001	40.93–67.94	7.26–8.41	339–378
WP	16.18–17.02	130.66–141.17	20.56–22.31	0.01–0.03	0.20–1.26	13.16–16.67	0.001–0.001	39.77–61.53	7.65–8.26	334–370
WB	15.31–17.02	129.92–150.38	20.64–22.36	0.02–0.07	0.00–0.20	13.11–16.78	0.001–0.006	42.51–67.38	7.51–8.21	352–372

2.2. Chemical and Physical Data

In total, 48 water samples were analyzed for their physical and chemical properties. They were collected in autumn (October 2017), spring (May 2018), and summer (August 2018) from 12 sampling points (four in each lake). All the sampling for physical and chemical properties was done in open water, with comparable environments selected as much as possible. Conductivity and pH were measured using a YSI 6600 V2 multiparameter sonde. Water for the chemical analyses was sampled 20–30 cm below the lake surface using 0.33 L polyethylene bottles, and stored in the dark at 4 °C, to limit ongoing chemical reactions. Ionic analyses were related to PO_4^{3-}, SO_4^{2-}, NO_3^-, F^-, CO_3^{2-}, Cl^-, and NO_2^-, as well as Na^+, Li^+, K^+, Mg^{2+}, NH_4^+, and Ca^{2+}. Laboratory measurement were performed using the Dionex ion chromatograph at the laboratory of the Institute of Nature Conservation, Polish Academy of Sciences. Spatial differences in the shorezone were calculated using the shorezone functionality index manual [38].

2.3. Diatom Data

In total, 48 samples were analyzed. They were collected in autumn (October 2017), spring (May 2018), and summer (August 2018) from 12 sampling points (four in each lake). Samples of periphyton were taken from the common reed, *Phragmites australis* (Cav.) Trin. ex Steud. Samples were cleaned by adding 37% H_2O_2, and were then heated. The reaction was completed by adding $KMnO_4$ and HCl. The cleaned diatom material dried on slides was mounted in Naphrax® synthetic resin. The slides were then analyzed using a Nikon Eclipse-80i microscope, with at least 400 diatom valves counted. Diatom identification mainly followed the procedures in [39–43]. Diatom indices (IOJ—diatom index for Polish lakes, Tj—trophic index, and GR—reference species index) were calculated using a program provided by the Polish Inspectorat of Environmental Protection (version 2010 and an updated version from May 2019) [24,25].

2.4. Statistical Data

2.4.1. β Diversity of Diatoms

Analyzed samples matrices were used to calculate β diversity [44]—(1) the total pairwise β diversity Sørensen dissimilarity index with partitioning (species turnover and nestedness) (betapart package R), and (2) local contribution to beta diversity (LCBD) with partitioning and species contribution to β diversity (SCBD) (adespatial package R). Data were cleaned before analysis—only species that had an abundance of at least 1% in 2 or more samples were used [33]—and were square-root-transformed to reduce the influence of very rare and abundant species on the diversity scores. In total, 6 β dissimilarity matrices were generated. Prior to the statistical analyses, all the abiotic variables were z-score-standardized (i.e., mean = 0, SD = 1) as well as log-transformed (if needed). Next, the chemical and physical variable distances among the samples were calculated (Euclidean distance). These matrices were transformed into a data frame and used for further analysis as distance-values. To quantify the association of each component of β diversity with the spatial and environmental factors, a multiple regression on the distance matrices ([45], MRM, Table 3) was used. To reduce the effect of spurious relationships between the variables, the MRM test was conducted with all the selected variables in the non-redundant variable sets. Then the non-significant variables from this initial MRM test were removed and the test was re-run. The significance of the partial regression was tested 999 times by a matrix permutation. The relationship between the β diversity indices in distance matrix and the selected variables was modelled using the random forest (RF) algorithm ([46] python 3.9 program—scikit learn 0.24 library). Optimal hyperparameters were found using a random search followed by a grid search (Table 4, with hyperparameters). The importance of each predictor variable (Table 4) was determined by: (1) permutation feature importance (rfpimp) [47], and (2) mean decrease accuracy (eli5) [48]. To reduce the effect of spurious relationships between variables, the RF model was developed with all the selected variables. Then, the variable(s) with the lowest contribution (less than 0.05) were removed and the model was re-developed until all contributed features were positive (add value) on the random forest model [47,48].

Table 3. β diversity models. 1: Redundancy analysis (RDA) for chosen β diversity index and its most important features: model variance constrained, model variance unconstrained, model probability, and probability for axis. 2: Variation partitioning and adjusted R^2 for analyzed factors. 3: Multiple regression on distance matrices (MDM)—probability for chosen factors and R^2. Adj—adjusted; R^2—coefficient of determination; p—probability; *—$p < 0.05$; **—$p < 0.01$.

Analysis	**Function**	**LCBD Total**	**LCBD Turnover**	**LCBD Nestedness**	**Beta Sørensen Total**	**Beta Sørensen Turnover**	**Beta Sørensen Nestedness**
1. Redundancy analysis	Model variance constrained	0.3763	0.4172	0.2580	1.056	0.3421	0.716
	Model variance uncontrained	4.6237	4.5828	4.742	3.944	4.6579	4.284
	Model p	0.011 *	0.006 **	0.048 *	0.001 **	0.017 *	0.001 **
	Axis p	0.008 **	0.006 **	0.048 *	0.002 **	0.012 *	0.002 **
2. Variation partitioning	Adj $R^2 SO_4^{2-}$	0.013	0.01842	−0.02015	0.16979	−0.02171	0.33897
	Adj $R^2 NO_3^-$	0.00721	−0.01534	0.09121	0.45749	0.13798	0.20454
	Adj $R^2 PO_4^{3-}$	0.04678	0.03172	0.01942	0.00153	−0.02019	0.00510
	Adj $R^2 NH_4^+$	−0.01761	−0.01066	−0.01324	0.04875	0.03053	−0.01343
3. Multiple regression on distance matrices	p SO_4^{2-}	-	-	-	-	-	-
	p NO_3^-	0.001 **	0.001 **	-	0.004 **	0.020 *	-
	p Ca^{2+}	-	-	-	0.001 **		0.001 **
	p Cl^-					0.001 **	-
	p PO_4^{3-}	0.032 *	0.018 *	-	-	-	-
	R^2	0.1085	0.1124	Not significant	0.2041	0.05953	0.1297

Table 4. β diversity models. 1: Random forest parameters of best model. 2 and 3: Importance analysis for chosen model (2: detailed information for rfpimp and 3: detailed information for eli5); OOB score—out-of-bag score. 4—Linear models (single values). Adj—adjusted; R^2—coefficient of determination; p—probability; *—$p < 0.05$; **—$p < 0.01$; ***—$p < 0.001$.

Analysis		**LCBD Total**	**LCBD Turnover**	**LCBD Nestedness**	**Beta Sørensen Total**	**Beta Sørensen Turnover**	**Beta Sørensen Nestedness**
1. Random forest —best parameters	Bootstrap	True	True	True	True	True	True
	Max depth	80	110	50	30	80	80
	Max features	3	Sqrt	3	2	2	2
	Min samples leaf	3	2	2	2	1	1
	Min samples split	5	2	2	4	4	4
	N estimators	600	400	200	100	200	200
	OOB score	True	True	True	True	True	True
2. Random forest feature importance (rfpimp)	SO_4^{2-}	0.305	0.232	0.128	0.419	0.241	0.140
	NO_3^-	0.266	0.201	0.122	0.229	0.236	0.293
	Ca^{2+}	0.130	0.146	0.562	0.403	0.176	0.400
	Cl^-	0.210	0.278	0.121	0.079	0.212	0.150
	NH_4^+	0.067	0.097	0.189	0.195	0.125	0.167
	PO_4^{3-}	0.083	0.106	0.083	0.062	0.104	0.098
	R^2	0.83	0.85	0.89	0.88	0.82	0.81
3. Random forest feature importance (eli5)	SO_4^{2-}	0.299	0.241	0.129	0.409	0.251	0.123
	NO_3^-	0.236	0.193	0.135	0.245	0.263	0.284
	Ca^{2+}	0.132	0.159	0.536	0.351	0.169	0.371
	Cl^-	0.210	0.320	0.147	0.0756	0.223	0.166
	NH_4^+	0.071	0.104	0.191	0.160	0.143	0.167
	PO_4^{3-}	0.098	0.148	0.096	0.055	0.100	0.91
	OOB score	0.52	0.42	0.55	0.44	0.31	0.28

Table 4. *Cont.*

Analysis		LCBD Total	LCBD Turnover	LCBD Nestedness	Beta Sørensen Total	Beta Sørensen Turnover	Beta Sørensen Nestedness
4. Linear model (single values)	p SO_4^{2-}	0.0020 **	0.000811 ***	-	0.0311 *	-	0.00000005 ***
	p NO_3^-	0.000549 ***	0.002330 **	0.0204 *	0.00000126 ***	0.00541 **	0.00000126 ***
	p Ca^{2+}	0.000284 ***	0.000240 ***	-	-	-	0.0000852 ***
	p Cl^-	-	-	-	-	-	-
	p PO_4^{3-}	-	-	-	-	-	-
	p NH_4^+	0.0399 *	0.044987 *	-	-	-	0.0123 *
	Adj R^2	0.26	0.257	0.0921	0.4992	0.138	0.597

To examine the changes in β diversity across selected environmental factors, we divided the samples into groups, which were nested in lake factors and season factors, and for each sampling point in those nested conditions, arithmetic means were calculated. In this way, we generated a single-point matrix, taking into consideration that single-point measurements are more powerful and recommended for calculations [44]. The statistical dependence between the explanatory variables was assessed using Pearson's correlation analysis and the variables with high correlation coefficients (Pearson $R^2 > 0.7$) were excluded from the models. Then, the relationships between the environmental factors and β diversity metrics were explored with linear models. We performed variation partitioning analyses as well as an RDA analysis (redundancy analysis [49–51], Table 3) to check which of the β diversity indices better corresponded with chemical and physical water features.

2.4.2. Seasonal Differences, Between-Lake Differences, and Effectiveness of Indices

The differences between seasons were analyzed in 3 datasets: (a) diatom assemblages, (b) diatom indices, and (c) an assessment based on diatom index. To assess differences in diatom assemblages, a nonmetric multidimensional scaling analysis was conducted (NMDS, [52]) based on the Bray-Curtis similarity matrix ([53], R 4.0.2 software, packages: vegan, plot3D). Data were cleaned before the analysis—only species which had an abundance of at least 1% in 2 or more samples were used [33]—and were square-root-transformed to reduce the influence of very rare and abundant species on ordination scores. In order to analyze the variability in the studied lakes, the datasets from the four lakes were ordered in a bidimensional space using the above-mentioned NMDS with Bray-Curtis as a dissimilarity measure [32,33]. Significant differences between the centroids of the multivariate areas covered by the four lakes were tested with PERMANOVA (permutational analysis of variance [54]) and interpreted as indicative of differences in the mean composition of diatom assemblages, diatom indices (IOJ), and assessments based on diatom indices among lakes (assessment based on IOJ) (R software, vegan package). PERMANOVA was performed to assess statistical differences between seasons and lakes: in diatom assemblages (Euclidean distance matrix [32] and parallel to the methods of [33] using NMDS), and in diatom indices and assessments based on a diatom index (R software, vegan package, pairwise adonis package). A test of the homogeneity of multivariate dispersion [55] was also applied to assess whether cells (groups of points nested in lake and season) (batadisper, R, vegan) had a similar dispersion (homogeneity of variance—equivalent to Levene's test). A correlation analysis was performed as the network analysis in JASP (Jeffrey's Amazing Statistics Program) [56,57] on the botnet package in R statistical software. The line thickness among stations reflected the correlation value (only significant correlations were represented); blue was positive, and red was negative. The nodes were positioned using the Fruchterman-Reingold algorithm, which organizes the network based on the strength of the connections between nodes. JASP network graphs were constructed on the base of each revealed species abundance, number of bioindicators, and environmental data in each lake and for each of the four seasons.

We used RDA ([49–51], R software, vegan package) to assess which of the diatom indices explained more of variance in the environmental dataset. Prior to reducing skewness and normalizing distributions for the data analyses, all physical and chemical variables were log-transformed when necessary, and all variables were normalized. Data were cleaned to avoid multicollinearity—the most correlated variables were excluded from analysis [32]. Characteristic species for every lake were revealed by IndVal [58].

2.5. Spatial Diversity

The representativeness of the samples was investigated by the comparison of original samples with Monte Carlo-simulated assemblages [32,33,51]. In order to simulate assemblages, specimens from the same lake in the same season were pooled and 1000 new samples for each were drawn at random (400 specimens per sample) [32,47,51]. Data sets were square-root-transformed and a PCoA (principal coordinate analysis [59]) using Bray-Curtis as a dissimilarity measurement was performed on each randomized dataset separately (R, package stats) and then plotted. The distribution was used to detect the representativeness of original species assemblages. Spatial within-lake variability was also assessed through comparing the Euclidean distance of original assemblages to the centroid of generated assemblages (simulated mean assemblages in PCoA). Boxplots were drawn to visualize differences. Samples with a distance to centroid that were located in the tail representing less than 5% were interpreted as poorly representative [51]. For comparison, there was a plotted cumulative PCoA plot for all simulated samples, with real samples highlighted. Simulated assemblages were also used for the calculation of the generated diatom index for Polish lakes (IOJ) (version 2010 and 2019) and assessments based on the diatom index. Distributions were visualized on histograms and on box-plots by comparing the Euclidean distance between the original dataset and the mean. General linear models were used to assess statistical differences between diatom assemblages and diatom indices in seasons and lakes. Samples with IOJ that were placed in the histogram tails representing less than 2.5% were interpreted as poorly representative.

3. Results

3.1. β Diversity

β diversity was mostly consistent with the turnover partition (mean—81%, median—85%), and nestedness on average had a 19% share (median—15%). Local contributions to the β diversity were mostly due to turnover partition (mean—88%, median—92%), which were higher than the nestedness (mean 11%, median—7%). The RDA showed a significant relationship between the environmental features for all β diversity indices, although with low explanation power (Table 3). The variation partitioning (Table 3) analysis gave the highest explanation power to NO_3^- and SO_4^{2-} for β Sørensen indices (β Sørensen total adjusted R^2 for NO_3^-—0.46, for SO_4^{2-}—0.17; β Sørensen turnover adjusted R^2 for NO_3^-—0.14; β Sørensen nestedness adj R^2 for NO_3^-—0.20, for SO_4^{2-}—0.34). The LCBD showed lower variation explanation, and PO_4^{3-} had the most significant contribution. The MRM analysis (Table 3) showed NO_3^- as a significant contributor to the LCBD and β Sørensen, but the explanation power was not noticed for LCBD nestedness. β Sørensen nestedness showed a correlation only with Ca^{2+} ions. The explanation powers for all distances (in MRM) were much lower than for the raw material analysis (random forest and linear models). Random forest (Table 4) showed the greatest contribution to the variance for NO_3^-, Cl^-, and SO_4^{2-} to β Sørensen as well as LCBD; similar results were noticed for turnover. However, for nestedness, the components Ca^{2+} and NH_4^+ had the greatest contribution. Linear models (Table 4) showed higher R^2: 0.26 for LCBD and 0.50 for β Sørensen. SO_4^{2-} and NO_3^- had the highest contribution to both β Sørensen and LCBD. The turnover was the most significant process, as shown for both the LCBD and β Sørensen indices. For SCBD, the most significant species were *Achnanthidium affinis*, *Brachysira neoexilis*, and *Cymbella affiniformis*. The most characteristic taxa for the analyzed lakes were: BW—*Gomphonema procerum*, *Encyonema ventricosum*, and *Nitzschia lacuum*; OK—*Nitzschia palea*; K—*Brachysira neoexilis*, *B. procera*, and *Eunotia arcubus*; Wigry—*Cymbella excisa*, *Fragilaria subconstricta*, and *Cocconeis placentula* (Table 5). The samples included mostly benthic diatoms, with low numbers of plankto-benthic species. The present diatom indicator taxa have mostly temperate temperature preferences (e.g., *Cymbella cymbiformis*) with the addition of eurythermic taxa. Oxygen (Figure 2) indicators (e.g., *Gomphonema vibrio*) showed high seasonal differences, with the highest contribution of well-oxygenated water in the spring and the lowest in autumn. Salinity (e.g., halophobes—*Cymbella proxima*) (Figure 3) and pH indicators (e.g., alkaliphiles—*Rhopalodia gibba*) showed low differences across samples. The lowest amount of saproxenes (e.g., *Delicata delicatula*) and highest of saprophiles (e.g., *Nitzschia palea*) were observed in O lake. The lowest results were also observed in O lake for the trophic state (Figure 4) (e.g., nitrogen autotrophic—*Gomphonema vibrio*).

Table 5. Indicator value for analyzed lakes. Indicator species values obtained by the indicator species analysis IndVal, p—probability.

	Lakes	Taxon 1	Indicator Value	*p*	Taxon 2	Indicator Value	*p*	Taxon 3	Indicator Value	*p*
1.	O	*Nitzschia palea*	0.31	0.025	-	-	-	-	-	-
2.	BW	*Gomphonema procerum*	0.52	0.001	*Encyonema ventricosum*	0.46	0.002	*Nitzschia lacuum*	0.39	0.001
3.	K	*Brachysira neoexilis*	0.45	0.001	*Brachysira procera*	0.44	0.03	*Eunotia arcubus*	0.43	0.004
4.	W	*Cymbella excisa*	0.65	0.001	*Fragilaria subconstricta*	0.51	0.002	*Cocconeis placentula*	0.41	0.006

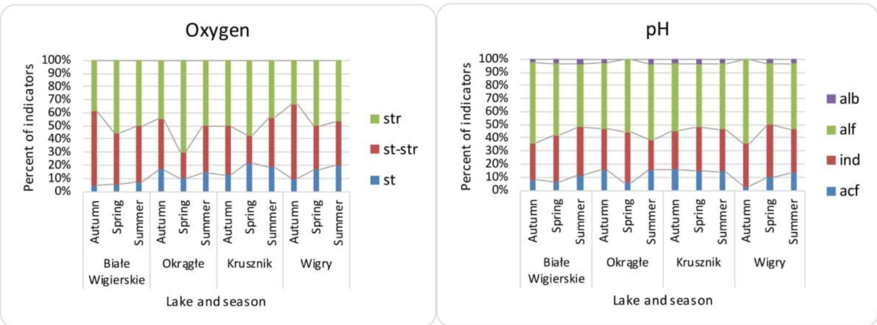

Figure 2. Distribution of diatom indicator taxa for oxygen and pH preferences in the Wigry lakes over the seasons. Oxygen (oxygenation and streaming) abbreviations: st—standing, low oxygenated water; str—streaming, well oxygenated water; st–str—low streaming, middle oxygenated water. pH abbreviations: alb—alkalibiontes; alf—alkaliphiles; ind—indifferents; acf—acidophiles. O—Okrągłe lake; BW—Białe Wigierskie lake; K—Krusznik lake; W—Wigry lake. Data only for indicator taxa.

3.2. Seasonal Differences, Differences between Lakes, and Effectiveness of Indices

The diversity of the studied lakes is included in the values of physical and chemical parameters (Table 2). The conductivity ranged between 131 and 378 µS/cm; in general, WK and WB had the highest values, and BW had the lowest. A JASP comparison of the similarity of diatom assemblages, ecological indicators, and environmental data from the studied lakes is presented in Figures 5 and 6. The JASP network plot shows that the BW assemblages were most similar (significant, $p < 0.05$); they formed one core with some of the K samples. The samples from other lakes were mostly divided into two cores (Figure 6). Season was an important differentiator for samples (Figure 6). The summer samples formed a distinctive core of similarity; the autumn and spring samples were more similar to each other. Overall, JASP showed that the analyzed lakes belonged, to some extent, to the same region with common climatic and landscape parameters. The most distinctive lake was W, as its samples were spread across clusters with a low correlation to each other.

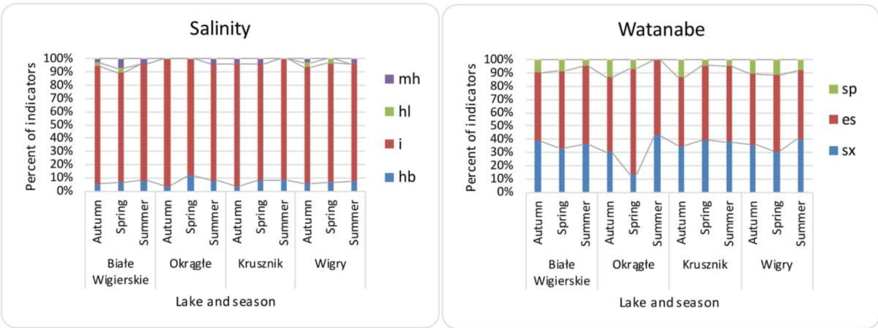

Figure 3. Distribution of diatom indicator taxa for salinity and Watanabe organic pollution in the Wigry lake over the seasons. Salinity (halobity group) abbreviations: i—indifferent oligohalobes; hl—halophiles; hb—halophobes; mh—mesohalobes. Watanabe (organic pollution indicators according to Watanabe) abbreviations: sx—saproxenes; es—eurysaprobes; sp—saprophiles. O—Okrągłe lake; BW—Białe Wigierskie lake; K—Krusznik lake; W—Wigry lake. Data only for indicator taxa.

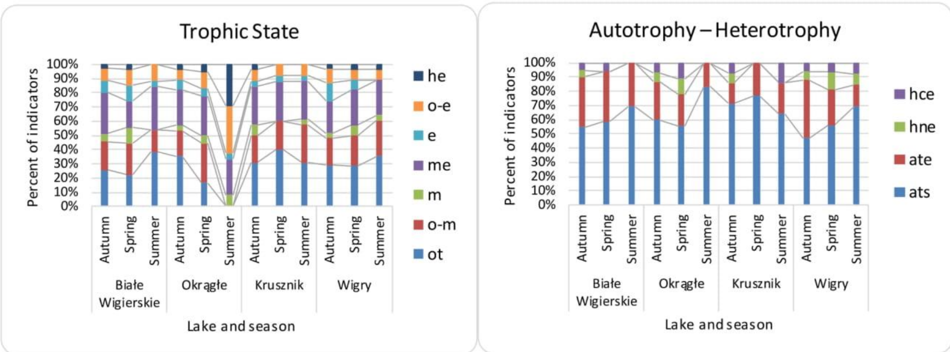

Figure 4. Distribution of diatom indicator taxa of trophic state and nutrition in the Wigry lakes over the seasons. Trophic state abbreviations: ot—oligotraphentic; o-m—oligo-mesotraphentic; m—mesotraphentic; me—meso-eutraphentic; e—eutraphentic; o-e—oligo-eutraphentic; he—hypereutraphentic. Autotrophy-heterotrophy (nitrogen uptake metabolism) abbreviations: ats—nitrogen-autotrophic taxa, tolerating very small concentrations of organically bound nitrogen; ate—nitrogen-autotrophic taxa, tolerating elevated concentrations of organically bound nitrogen; hne—facultatively nitrogen-heterotrophic taxa, needing periodically elevated concentrations of organically bound nitrogen; hce—facultatively nitrogen-heterotrophic taxa, needing elevated concentrations of organically bound nitrogen. O—Okrągłe lake; BW—Białe Wigierskie lake; K—Krusznik lake; W—Wigry lake. Data only for indicator taxa.

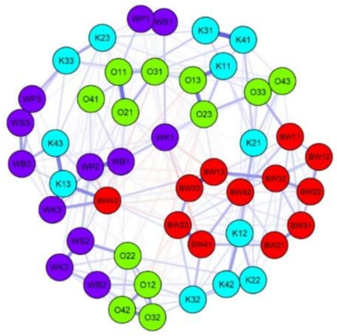

Figure 5. Jeffrey's Amazing Statistics Program (JASP) network plot of correlation on a level of more than 50% (significant only) for species diversity, ecological indicators, and environmental data of the lakes in the Wigry National Park. O—Okrągłe lake; BW—Białe Wigierskie lake; K—Krusznik lake; WK—Wigry lake Krzyżańska bay; WS—Wigry lake Słupiańska bay; WP—Wigry lake Piaski; WB—Wigry lake Bryzgiel. The second character is a code for sample (1–4), and the second number is a code for season: 1—autumn, 2—spring, 3—summer.

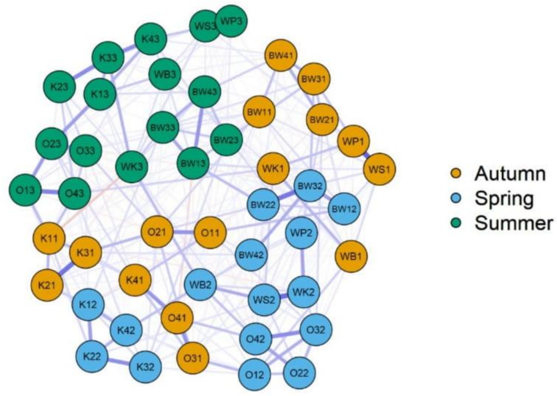

Figure 6. Jeffrey's Amazing Statistics Program (JASP) network plot of correlation on the level more than 50% (significant only) for seasonal species diversity, ecological indicators, and environmental data of the lakes in the Wigry National Park. O—Okrągłe lake; BW—Białe Wigierskie lake; K—Krusznik lake; WK—Wigry lake Krzyżańska bay; WS—Wigry lake Słupiańska bay; WP—Wigry lake Piaski; WB—Wigry lake Bryzgiel. The second character is a code for sample (1–4), and the second number is a code for season: 1—autumn, 2—spring, 3—summer.

The most abundant species was *Achnanthidium minutissimum*, and it was dominant within most samples (87% of samples had more than 10% abundance; the range was 5–88%). An NMDS on the cleared abundance data with 1000 permutations was performed. As the stress in the 2D analysis was above 0.2, the NMDS was re-run in 3D (stress reached—0.16). A dissimilarity matrix obtained from the NMDS was used to analyze the differences between lakes and seasons (this procedure was chosen as the most suitable for data in studies such as [32,33]). A PERMANOVA global test showed significant differences in the abundance data between seasons and lakes (pseudo F = 9.6392, $p < 0.001$ for lakes; pseudo F = 7.6568, $p < 0.001$ for seasons). All four lakes and three seasons were significantly different ($p < 0.05$). The IOJ, version 2010, ranged between 0.688 for WP and 0.939 for BW, and the version from 2019 ranged between 0.66 for WP and 0.94 for BW. The class of water quality and index of saprobity showed mostly second-class indicators (Figure 7). The PERMANOVA test showed significant differences in the IOJ between lakes and seasons for IOJ versions 2010 and 2019 (for 2010: pseudo F = 3.9, $p < 0.01$ for lakes, and pseudo F = 4.05, $p < 0.05$ for seasons; for 2019: pseudo F = 3.9, $p < 0.02$ for lakes). Pairwise tests showed significant differences for 2010 between K and BW, K and W, summer and autumn, and autumn and spring; for 2019 between K and BW and between K and W, with no significant differences between seasons.

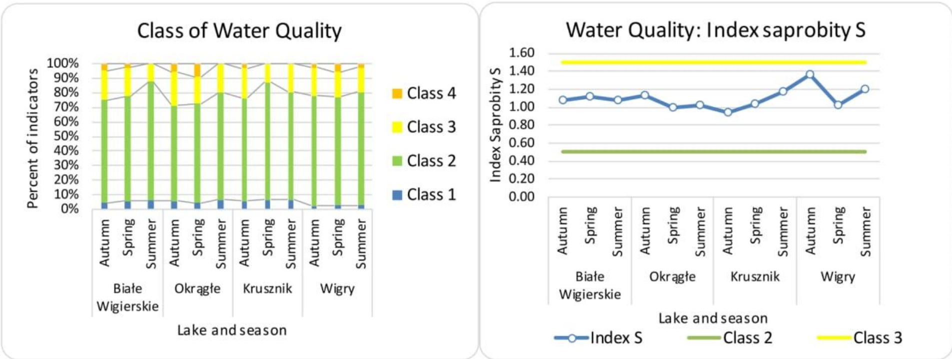

Figure 7. Class of water quality—distribution of diatom indicator taxa of water quality class (in EU color code). Water quality—average index saprobity in the Wigry lakes over seasons. O—Okrągłe lake; BW—Białe Wigierskie lake; K—Krusznik lake; W—Wigry lake. Data only for indicator taxa.

PERMANOVA showed differences in the assessments between lakes for the 2010 version (pseudo F = 7.8571, $p < 0.002$), but non-significant differences between seasons for the 2010 version and between lakes and seasons for the 2019 version. PERMANOVA showed non-significant differences between IOJ 2010 and IOJ 2019 and non-significant differences for the assessments.

We used an RDA to assess which of the diatom indices explained more of the variance in the environmental dataset. The RDAs used to model the variation in cleared and normalized nutrient levels, compared to the diatom indices and assessments used as predictors, showed significant explanation power for version 2010 (variance = 0.54, residuals = 8.47, $p = 0.032$, paxis = 0.038) and non-significant explanation power for IOJ version 2019 and both assessments.

3.3. Spatial Diversity

Monte Carlo-simulated assemblages were plotted using PCoA; real samples were highlighted (Figure 8). The centroids generated in the PCoA for each separately simulated data set were interpreted as mean assemblages and used to calculate the distances to this assembly. GLM tests were performed to compare the representativeness of samples for lakes and seasons; the results showed significant differences between lakes (BW-K $p = 0.015$, W-K $p = 0.018$) and no significant differences between seasons (Figure 9). A total of 79% of samples were poorly representative of the mean assemblages for lakes; however, most of the distances were less than 0.3, which indicated good representation [32,60].

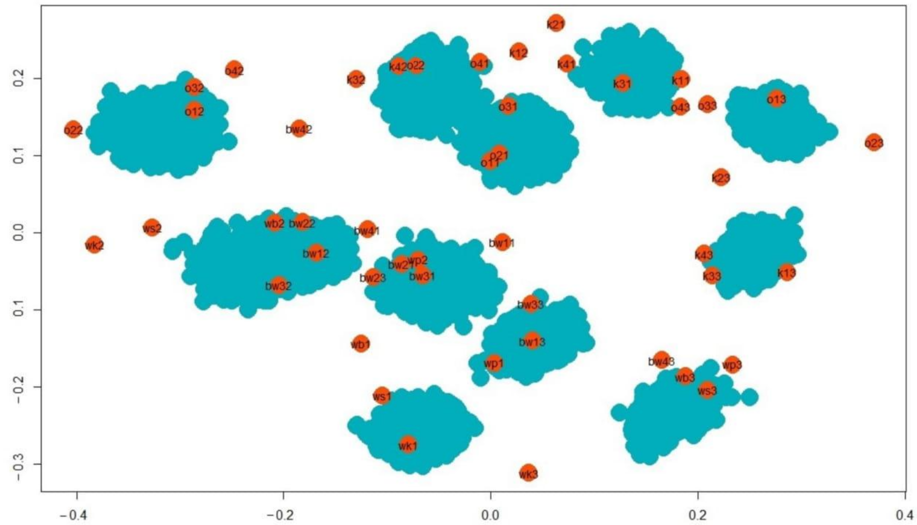

Figure 8. Principal coordinate analysis (PCoA) cumulative plot; red dots—real samples and blue dots—generated samples. Letters are code for lakes; first number is the code for the sample, and second number is the code for season: 1—autumn, 2—spring, 3—summer.

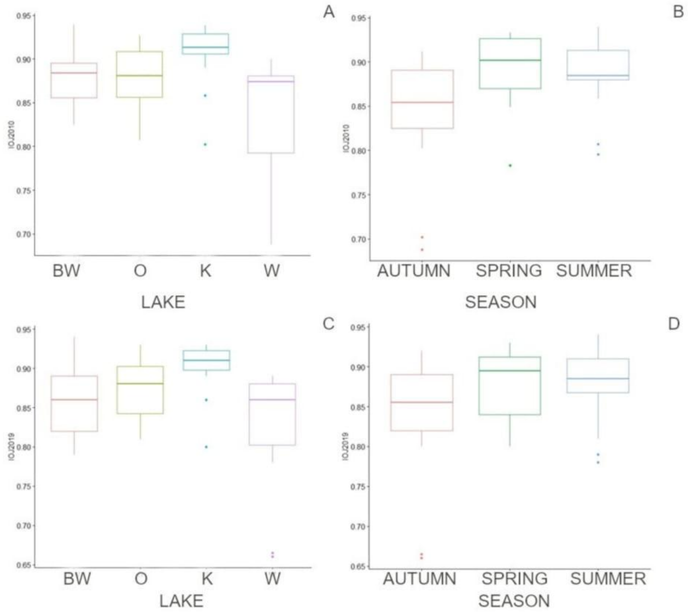

Figure 9. General linear model (GLM) analysis for groups—lakes (**A**,**B**) and seasons (**C**,**D**); IOJ 2010 (**A**,**C**), IOJ 2019 (**B**,**D**). O—Okrągłe lake; BW—Białe Wigierskie lake; K—Krusznik lake; W—Wigry lake.

The generated assemblages were used to calculate the IOJ version 2010 and 2019, histograms were used to plot the differences, and the real samples were highlighted. A total of 69% of the real samples were poorly representative of the IOJ of the mean assemblages for lakes for version 2010, and 73% for version 2019.

4. Discussion

4.1. β Diversity

Studies using variation partitioning to unravel metacommunity mechanisms assume that, in general, (I) species-sorting—if solely the "environmental variables" fraction—significantly explains the community structures; (II) neutral theory or patch dynamics—if only the "spatial variables" fraction—is significant; and (III) the mass-effect concept or the combination of species-sorting and mass-effect, if both fractions have significant explanatory power [61]. In our case, β diversity was mainly constructed with species turnover and was highly driven by environmental changes, with no statistically significant changes in the spatial factors represented by SFI; the filtering effect of the lakes' local environmental characteristics and species sorting played a significant role (Tables 3 and 4). Our results correspond well with the data presented by Epele et al. [62]. The variables with the most explanation power were NO^{3-} and $SO_4{}^{2-}$, both being indicators of water eutrophication and human impact on the environment [63]. Higher concentrations as well as differences in NO^{3-} and $SO_4{}^{2-}$ resulted in the highest species turnover as well as species nestedness; however, the turnover contribution was more powerful in the tested variance. The LCBD showed a lower variation explanation than the β Sørensen indices. These findings are consistent with research where LCBD β diversity was not well-determined by the local environmental characteristics [3]. A high dominance in the studied lakes contributed to lower LCBD values in comparison to the presence-absence β Sørensen indices. The distance analysis was less powerful than the single-value analysis, which is consistent with [44]. However, in our case random forest successfully showed similar patterns for distance matrices, similarly to the linear regression model.

The species with the highest incidence based on SCBD were *Achnanthidium affinis*, *Brachysira neoexilis*, and *Cymbella affiniformis*. These taxa are relatively common; however, they are not the most abundant ones. This score is comparable to the results shown by Szabó et al. [3]. The most common taxa are present in most of the samples, so they have little effect on the β diversity; on the other hand, species that are relatively common have high turnover, which contributes to overall β diversity. This mechanism is consistent with our findings that the most common taxa did not contribute to β diversity, and as well, they skewed the indication analysis. The presence-absence data underweighted those taxa so that the overall explanation power rose in the β Sørensen indices. Higher LCBD values were seen in samples with high or low species richness [1]. The high LCBD index could be a result of special ecological conditions, which should be given more attention in terms of conservation [8].

4.2. Spatial and Seasonal Differences

Seasonal differences between diatom assemblages were reported numerous times; however, whether these differences were caught by ecological assessments remains mostly unknown, especially for the diatom index for Polish lakes. Our results show significant differences in the qualitative composition of diatom assemblages occurring in individual seasons and in the diatom index for Polish lakes (IOJ) (continuous variable, 0–1); however, an ecological assessment performed using IOJ 2010and 2019 (very good, good, moderate, poor, bad) showed no significant differences between the seasons. More samples per year gave us redundant information about the ecological status (very good, good, moderate, poor, bad) despite significant differences between assemblages and the IOJ itself (continuous variable, 0–1). Elias et al. [31] reported similar results for streams and IPS assessments. Diatoms are naturally spatially and seasonally dynamic [31,33]. However, many species exhibit a similar response to the environment, and despite taxonomical differences, assessments remain comparable. The spatial differences within lakes were mostly lower than a distance of 0.3 to the centroids based on a PCoA performed on Monte Carlo-generated assemblages [32,59]. If the average composition of diatom assemblages significantly differs between two lakes with contrasting environmental characteristics, then single samples can be faithfully used for environmental bioindication, provided the environmental variable of interest explains a significant portion of the diatom variance [32]. If that is not the case, and the diatom composition between-lake variance is lower than the within-lake variance, then the number of single samples needed to represent the environmental differences among lakes is highly dependent on the sampling design [32,51]. In our analysis, the between-lake variance was far greater than the within-lake variance. These results indicate a good representation of lake samples. The IOJ ecological assessments mostly were not differentiated within a lake, despite the fact that over 65% of the IOJs (continuous variable, 0–1) calculated for real samples were considered as poor representations of the lake. The border lines between the ecological assessment groups were so wide that different IOJs gave similar assessments. More samples per lake and more samples per year seemed to give redundant information about the ecological status, especially for current assessment borders. Prygiel et al. [64] and Kelly et al. [10] have suggested that the uncertainty due to the sampling process itself is relatively small in comparison to other sources, such as, for example, taxonomic issues. Kelly et al. [10] have suggested that more reliable information would be given by more samples over a longer period (not seasonally, but in different hydrological years). Rimet et al. [34] and Marzin et al. [65] reported for their studies that spatial factors play a very limited role; on the other hand, water currents, animals, winds, and humans play intense roles in the diversity of diatoms at regional scales.

Diatoms have been used to assess the ecological status of lakes for years; however, from the early stages of implementing diatom indices in lakes to the current time, researchers have reported problems with the most abundant species such as *Achnanthidium minutissimum* [66,67]. A dominance of *A. minutissimum* is reported in many lakes, such as deep lakes with low to medium alkalinity [63]. *Achnanthidium minutissimum* is described as a pioneer species, so small disturbances such as water fluctuation, wind, or grazing will favor this species. However, *A. minutissimum* is a dominant species even in water reservoirs that have been severely impacted by metal contamination [68]. On the other hand, more severe disturbance, which impact grazing organisms, favor a more even structure of the assemblages [69]. Kelly et al. [16] showed that this process occurs on the moderate-to-good border of ecological assessments, and that diatom richness gives a unimodal response. However, we did not observe that this kind of assemblage difference caused such differences in the ecological assessment based on the IOJ. Moreover, we observed that less affected, mesotrophic lakes had a more even structure [70] compared to eutrophic lakes with high dominance, similarly to what has been reported by Stenger-Kovács et al. [71].

Some researchers have evaluated indices by checking how two indices react to differences in water chemistry (e.g., [72]). We evaluated two indices: version 2010 IOJ, and the updated version 2019. As we found, the grouping of hard-to-identify species into complexes using the updated 2019 version of IOJ gave us non-significant relationships with water chemistry, in comparison to significant relationships between the physical and chemical variables of water using the 2010 version. This is consistent with the findings of other research groups [73], as more taxonomic effort gives better results in terms of an indication of water chemistry changes. This aspect is important, especially for countries implementing ecological assessments and for those looking to simplify environmental inspections. We suggest that there is a high risk of losing useful data through this simplification. However, it is worth mentioning that both versions of IOJ had a low capacity for detecting environmental changes ($R^2 < 0.25$, [74]).

Diatoms have proven to be useful indicators of water quality. Because of their short generation time, they give information in short time frames [18]. It is useful to collect such fast-paced changes in the environment. These abilities show that phytobenthos and macrophytes, even though they are both producers, do not give redundant information and should be used in parallel. A healthy ecosystem is a good indicator that a water body is being exploited in sustainable manner [75]. The results of the calculations of the diatom index for Polish lakes in recent years have not been well-received [21–23]. However, in this shape and form, the IOJ does not indicate problems with an ecosystem [22]. Unfortunately, more spatial and seasonal samples are not the answer we are looking for. An analysis of all the phytobenthos groups (as performed by Kelly in [69]), exclusion of the most dominant species (as performed by Szczepocka et al. in [67]), assembling species into functional groups, or other approaches [33,76] are possibly the next options to consider. More taxonomic effort has proven to increase the indication accuracy [73]; however, due to hardware and human resource shortages in provincial inspectorates of environmental protection, we do not recommend it.

5. Conclusions

The β diversity of diatoms and their local contributions can be explained by the effects of environmental variables—a total of 49.9% and 26.0% of diatom variance was explained respectively ($p < 0.01$) with no significant effect of spatial differences represented by SFI. In our analysis, we found that both versions of IOJ (2010 and 2019) had a low capacity for detecting environmental changes. The grouping of hard-to-identify species into complexes in the updated version of IOJ 2019, which is useful for practical reasons, gave us a non-significant relationship for water chemistry, in comparison to a significant relationship between the physical and chemical variables of water by using the 2010 version. As real samples and Monte Carlo-generated samples were mostly at a PCoA distance of 0.3 from the centroids, one sample per lake and per year seems sufficient and is a compromise between sampling effort and financial cost, especially taking into consideration resampling in longer periods of time (i.e., once a year). However, we recommend changes to the IOJ itself, due to its low explanation power and low sensitivity to environmental changes. Species with high dominance in lakes lower the indication capabilities of IOJ as well as the percentage contribution to β indices (LCBD). Our findings provide insights into the mechanisms responsible for community organization along environmental gradients from the perspective of β diversity components and the mechanisms of indication values of diatoms for lakes, and can be used especially by countries implementing ecological assessments.

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References

1. Wu, K.; Zhao, W.; Li, M.; Picazo, F.; Soininen, J.; Shen, J.; Zhu, L.; Cheng, X.; Wang, J. Taxonomic Dependency of Beta Diversity Components in Benthic Communities of Bacteria, Diatoms and Chironomids along a Water-Depth Gradient. *Sci. Total Environ.* **2020**, *741*, 140462. [CrossRef] [PubMed]
2. Maloufi, S.; Catherine, A.; Mouillot, D.; Louvard, C.; Couté, A.; Bernard, C.; Troussellier, M. Environmental Heterogeneity among Lakes Promotes Hyper β-Diversity across Phytoplankton Communities. *Freshw. Biol.* **2016**, *61*, 633–645. [CrossRef]
3. Szabó, B.; Lengyel, E.; Padisák, J.; Stenger-Kovács, C. Benthic Diatom Metacommunity across Small Freshwater Lakes: Driving Mechanisms, β-Diversity and Ecological Uniqueness. *Hydrobiologia* **2019**, *828*, 183–198. [CrossRef]
4. Baselga, A.; Orme, C.D.L. Betapart: An R Package for the Study of Beta Diversity. *Methods Ecol. Evol.* **2012**, *3*, 808–812. [CrossRef]
5. Baselga, A. Partitioning the Turnover and Nestedness Components of Beta Diversity: Partitioning Beta Diversity. *Glob. Ecol. Biogeogr.* **2010**, *19*, 134–143. [CrossRef]
6. Legendre, P.; De Cáceres, M. Beta Diversity as the Variance of Community Data: Dissimilarity Coefficients and Partitioning. *Ecol. Lett.* **2013**, *16*, 951–963. [CrossRef]
7. Specziár, A.; Árva, D.; Tóth, M.; Móra, A.; Schmera, D.; Várbíró, G.; Erős, T. Environmental and Spatial Drivers of Beta Diversity Components of Chironomid Metacommunities in Contrasting Freshwater Systems. *Hydrobiologia* **2018**, *819*, 123–143. [CrossRef]
8. Legendre, P. Interpreting the Replacement and Richness Difference Components of Beta Diversity: Replacement and Richness Difference Components. *Glob. Ecol. Biogeogr.* **2014**, *23*, 1324–1334. [CrossRef]
9. Nõges, P.; Poikane, S.; Cardoso, A.C.; van de Bund, W. Water Framework Directive the Way to Water Ecosystems Sustainability in Europe. *LakeLine* **2006**, *21*, 36–43.
10. Kelly, M.; Bennion, H.; Burgess, A.; Ellis, J.; Juggins, S.; Guthrie, R.; Jamieson, J.; Adriaenssens, V.; Yallop, M. Uncertainty in Ecological Status Assessments of Lakes and Rivers Using Diatoms. *Hydrobiologia* **2009**, *633*, 5–15. [CrossRef]
11. Dam, H.; Mertens, A.; Sinkeldam, J. A Coded Checklist and Ecological Indicator Values of Freshwater Diatoms from The Netherlands. *Neth. J. Aquat. Ecol.* **1994**, *28*, 117–133. [CrossRef]
12. Hofmann, G.; Werum, M.; Lange-Bertalot, H.; Lange-Bertalot, H. *Diatomeen im Süßwasser-Benthos von Mitteleuropa: Bestimmungsflora Kieselalgen für die Ökologische Praxis; über 700 der Häufigsten Arten und Ihre Ökologie*; Gantner: Ruggell, Austria, 2011.
13. Marra, R.C.; Algarte, V.M.; Ludwig, T.A.V.; Padial, A.A. Diatom Diversity at Multiple Scales in Urban Reservoirs in Southern Brazil Reveals the Likely Role of Trophic State. *Limnologica* **2018**, *70*, 49–57. [CrossRef]
14. Cristóbal, G.; Blanco, S.; Bueno, G. (Eds.) *Modern Trends in Diatom Identification: Fundamentals and Applications*; Developments in Applied Phycology; Springer: Cham, Switzerland, 2020.
15. Birk, S.; Bonne, W.; Borja, A.; Brucet, S.; Courrat, A.; Poikane, S.; Solimini, A.; van de Bund, W.; Zampoukas, N.; Hering, D. Three Hundred Ways to Assess Europe's Surface Waters: An Almost Complete Overview of Biological Methods to Implement the Water Framework Directive. *Ecol. Indic.* **2012**, *18*, 31–41. [CrossRef]
16. Kelly, M.; Urbanic, G.; Acs, E.; Bennion, H.; Bertrin, V.; Burgess, A.; Denys, L.; Gottschalk, S.; Kahlert, M.; Karjalainen, S.M.; et al. Comparing Aspirations: Intercalibration of Ecological Status Concepts across European Lakes for Littoral Diatoms. *Hydrobiologia* **2014**, *734*, 125–141. [CrossRef]
17. Poikane, S.; Kelly, M.; Cantonati, M. Benthic Algal Assessment of Ecological Status in European Lakes and Rivers: Challenges and Opportunities. *Sci. Total Environ.* **2016**, *568*, 603–613. [CrossRef]
18. Schaumburg, J.; Schranz, C.; Foerster, J.; Gutowski, A.; Hofmann, G.; Meilinger, P.; Schneider, S.; Schmedtje, U. Ecological Classification of Macrophytes and Phytobenthos for Rivers in Germany According to the Water Framework Directive. *Limnologica* **2004**, *34*, 283–301. [CrossRef]
19. Poikane, S.; Birk, S.; Böhmer, J.; Carvalho, L.; de Hoyos, C.; Gassner, H.; Hellsten, S.; Kelly, M.; Lyche Solheim, A.; Olin, M.; et al. A Hitchhiker's Guide to European Lake Ecological Assessment and Intercalibration. *Ecol. Indic.* **2015**, *52*, 533–544. [CrossRef]
20. Kelly, M.G.; Chiriac, G.; Soare-Minea, A.; Hamchevici, C.; Juggins, S. Use of Phytobenthos to Evaluate Ecological Status in Lowland Romanian Lakes. *Limnologica* **2019**, *77*, 125682. [CrossRef]
21. Bielczyńska, A. Bioindication on the Basis of Benthic Diatoms: Advantages and Disadvantages of the Polish Phytobenthos Lake Assessment Method (IOJ—The Diatom Index for Lakes)/Bioindykacja Na Podstawie Okrzemek Bentosowych: Mocne i Słabe Strony Polskiej Metody Oceny Jezior Na Podstawie Fitobentosu (IOJ—Indeks Okrzemkowy Jezior). *Ochr. Śr. Zasobów Nat.* **2015**, *26*, 48–55. [CrossRef]
22. Ciecierska, H.; Kolada, A. ESMI: A Macrophyte Index for Assessing the Ecological Status of Lakes. *Environ. Monit. Assess.* **2014**, *186*, 5501–5517. [CrossRef]
23. Wiech, A.K.; Marciniewicz-Mykieta, M.; Toczko, B. *Stan Środowiska w Polsce. Raport 2018*; Główny Inspektorat Ochrony Środowiska: Warsaw, Poland, 2018.
24. Picińska-Fałtynowicz, J.; Błachuta, J. *Przewodnik Metodyczny. Zasady Poboru i Opracowania Prób Fitobentosu Okrzemkowego z Rzek i Jezior*; GIOŚ: Wrocław, Poland, 2010.
25. Zgrundo, A.; Peszek, Ł.; Poradowska, A. *Podręcznik Do Monitoringu i Oceny Rzecznych Jednolitych Części Wód Powierzchniowych Na Podstawie Fitobentosu*; OceanSense Maciej Fojcik: Gdańsk, Poland, 2018. (In Polish)
26. King, L.; Clarke, G.; Bennion, H.; Kelly, M.; Yallop, M. Recommendations for Sampling Littoral Diatoms in Lakes for Ecological Status Assessments. *J. Appl. Phycol.* **2006**, *18*, 15–25. [CrossRef]

27. Lodewijks, P.; Brouwers, J.; Van Hooste, H.; Meynaerts, E. *Energie-En Klimaatscenario's Voor de Sectoren Energie En Industrie*; MIRA 2009 Wetenschappelijk Rapport; Vlaamse Milieumaatschappij: Mechelen, Belgium, 2009; Available online: https://www.academia.edu/19642138/Energie_en_Klimaatscenario_s_voor_de_sectoren_Energie_en_Industrie (accessed on 1 June 2022).
28. Charles, D.F.; Kelly, M.G.; Stevenson, R.J.; Poikane, S.; Theroux, S.; Zgrundo, A.; Cantonati, M. Benthic Algae Assessments in the EU and the US: Striving for Consistency in the Face of Great Ecological Diversity. *Ecol. Indic.* **2021**, *121*, 107082. [CrossRef]
29. Picińska-Fałtynowicz, J.; Soszka, H. Fitobentos. In *Ocena Stanu Ekologicznego Wód Zlewni Rzeki Wel. Wytyczne Do Zintegrowanej Oceny Stanu Ekologicznego Rzek i Jezior na Potrzeby Planów Gospodarowania Wodami w Dorzeczu*; Wydawnictwo IRŚ: Olsztyn, Poland, 2011.
30. Gołdyn, R.; Messyasz, B. Ocena Stanu Ekologicznego Jeziora Durowskiego w Roku 2013; Wydział Biologii Uniwersytetu im. Adama Mickiewicza w Poznaniu: Poznań, Poland. 2013. Available online: https://www.wagrowiec.eu/assets/files/dzialy/ochrona_srodowiska/dokumenty-do-pobrania/ocena-stanu/2018-stan-ekologiczny-jeziora-durowskiego.pdf (accessed on 1 June 2022).
31. Elias, C.L.; Vieira, N.; Feio, M.J.; Almeida, S.F.P. Can Season Interfere with Diatom Ecological Quality Assessment? *Hydrobiologia* **2012**, *695*, 223–232. [CrossRef]
32. Hassan, G.S. Within versus Between-Lake Variability of Sedimentary Diatoms: The Role of Sampling Effort in Capturing Assemblage Composition in Environmentally Heterogeneous Shallow Lakes. *J. Paleolimnol.* **2018**, *60*, 525–541. [CrossRef]
33. Riato, L.; Leira, M. Heterogeneity of Epiphytic Diatoms in Shallow Lakes: Implications for Lake Monitoring. *Ecol. Indic.* **2020**, *111*, 105988. [CrossRef]
34. Rimet, F.; Bouchez, A.; Montuelle, B. Benthic Diatoms and Phytoplankton to Assess Nutrients in a Large Lake: Complementarity of Their Use in Lake Geneva (France-Switzerland). *Ecol. Indic.* **2015**, *53*, 231–239. [CrossRef]
35. Andrzejczyk, T.; Brzeziecki, B. The Structure and Dynamics of Old-Growth *Pinus sylvestris* (L.) Stands in the Wigry National Park, North-Eastern Poland. *Vegetatio* **1995**, *117*, 81–94. [CrossRef]
36. Drzymulska, D.; Zieliński, P. Phases and Interruptions in Postglacial Development of Humic Lake Margin (Lake Suchar Wielki, NE Poland). *Limnol. Rev.* **2014**, *14*, 13–20. [CrossRef]
37. Górniak, A. Current Climatic Conditions of Lake Regions in Poland and Impacts on Their Functioning. In *Polish River Basins and Lakes—Part I.*; Korzeniewska, E., Harnisz, M., Eds.; Springer International Publishing: Cham, Switzerland, 2020; Volume 86, pp. 1–25. [CrossRef]
38. Nowicka, B.; Nadolna, A. Shorezone Functionality Index—Lake Charzykowskie Case Study. In *Natural and Athropogenic Transformation of Lakes, Proceedings of the International Limnological Conference, Łagów Lubuski, Poland, 19–21 September 2012*; Institute of Meteorology and Water Management: Poznań, Poland, 2012.
39. Lange-Bertalot, H.; Hofmann, G.; Werum, M.; Cantonati, M.; Kelly, M. *Freshwater Benthic Diatoms of Central Europe: Over 800 Common Species Used in Ecological Assessment*; English edition with updated taxonomy and added species; Koeltz Botanical Books: Schmitten-Oberreifenberg, Germany, 2017.
40. Wojtal, A.Z.; Ector, L.; Van de Vijver, B.; Morales, E.A.; Blanco, S.; Piatek, J.; Smieja, A. The Achnanthidium Minutissimum Complex (Bacillariophyceae) in Southern Poland. *Algological Stud.* **2011**, *136–137*, 211–238. [CrossRef]
41. Lange-Bertalot, H.; Ulrich, S. Contributions to the Taxonomy of Needle-Shaped Fragilaria and Ulnaria Species. *Lauterbornia* **2014**, *78*, 1–73.
42. Van de Vijver, B. Analysis of the Type Material of Navicula Brachysira Brébisson with the Description of Brachysira Sandrae, a New Raphid Diatom (Bacillariophyceae) from Iles Kerguelen (TAAF, Sub-Antarctica, Southern Indian Ocean). *Phytotaxa* **2014**, *184*, 139. [CrossRef]
43. Delgado, C.; Novais, M.H.; Blanco, S.; de Almeida, S.F. Examination and Comparison of Fragilaria Candidagilae sp. nov. with Type Material of *Fragilaria Recapitellata*, *F. Capucina*, *F. Perminuta*, *F. Intermedia* and *F. Neointermedia* (*Fragilariales*, *Bacillariophyceae*). *Phytotaxa* **2015**, *231*, 1. [CrossRef]
44. Legendre, P.; Borcard, D.; Peres-Neto, P.R. Analyzing Beta Diversity: Partitioning the Spatial Variation of Community Composition Data. *Ecol. Monogr.* **2005**, *75*, 435–450. [CrossRef]
45. Lichstein, J. Multiple regression on distance matrices: A multivariate spatial analysis tool. *Plant Ecol.* **2006**, *188*, 117–131. [CrossRef]
46. Biau, G.; Scornet, E. A Random Forest Guided Tour. *TEST* **2016**, *25*, 197–227. [CrossRef]
47. Altmann, A.; Toloşi, L.; Sander, O.; Lengauer, T. Permutation Importance: A Corrected Feature Importance Measure. *Bioinformatics* **2010**, *26*, 1340–1347. [CrossRef] [PubMed]
48. Gómez-Ramírez, J.; Ávila-Villanueva, M.; Fernández-Blázquez, M. Selecting the Most Important Self-Assessed Features for Predicting Conversion to Mild Cognitive Impairment with Random Forest and Permutation-Based Methods. *Sci. Rep.* **2020**, *10*, 20630. [CrossRef]
49. Van den Wollenberg, A. Redundancy Analysis an Alternative for Canonical Correlation Analysis. *Psychometrika* **1977**, *42*, 207–219. [CrossRef]
50. Vilmi, A.; Karjalainen, S.M.; Landeiro, V.L.; Heino, J. Freshwater Diatoms as Environmental Indicators: Evaluating the Effects of Eutrophication Using Species Morphology and Biological Indices. *Environ. Monit. Assess.* **2015**, *187*, 243. [CrossRef]

51. Heggen, M.P.; Birks, H.H.; Heiri, O.; Grytnes, J.-A.; Birks, H.J.B. Are Fossil Assemblages in a Single Sediment Core from a Small Lake Representative of Total Deposition of Mite, Chironomid, and Plant Macrofossil Remains? *J. Paleolimnol.* **2012**, *48*, 669–691. [CrossRef]
52. Kenkel, N.; Orloci, L. Applying Metric and Nonmetric Multidimensional Scaling to Ecological Studies: Some New Results. *Ecology* **1986**, *67*, 919–928. [CrossRef]
53. Beals, E. Bray-Curtis Ordination: An Effective Strategy for Analysis of Multivariate Ecological Data. In *Advances in Ecological Research*; Academic Press: Cambridge, MA, USA, 1984; pp. 1–55. [CrossRef]
54. Anderson, M. *Permutational Multivariate Analysis of Variance (PERMANOVA)*; Balakrishnan, N., Colton, T., Everitt, B., Piegorsch, W., Ruggeri, F., Teugels, J.L., Eds.; John Wiley & Sons: Hoboken, NJ, USA, 2017; pp. 1–15. [CrossRef]
55. Anderson, M. Distance-Based Tests for Homogeneity of Multivariate Dispersions. *Biometrics* **2005**, *62*, 245–253. [CrossRef] [PubMed]
56. Love, J.; Selker, R.; Marsman, M.; Jamil, T.; Dropmann, D.; Verhagen, J.; Ly, A.; Gronau, Q.F.; Smíra, M.; Epskamp, S.; et al. JASP: Graphical Statistical Software for Common Statistical Designs. *J. Stat. Soft.* **2019**, *88*, 1–17. [CrossRef]
57. Krupa, E.; Barinova, S.; Romanova, S.; Aubakirova, M.; Ainabaeva, N. Planktonic Invertebrates in the Assessment of Long-Term Change in Water Quality of the Sorbulak Wastewater Disposal System (Kazakhstan). *Water* **2020**, *12*, 3409. [CrossRef]
58. Dufrêne, M.; Legendre, P. Species assemblages and indicator species: The need for a flexible asymmetrical approach. *Ecol. Monogr.* **1997**, *67*, 345–366. [CrossRef]
59. Zuur, A.; Ieno, E.; Smith, G. Principal coordinate analysis and non-metric multidimensional scaling. In *Analysing Ecological Data*; Statistics for Biology and Health; Springer Science: New York, NY, USA, 2007; pp. 259–264. [CrossRef]
60. Kelly, M.G. Use of Similarity Measures for Quality Control of Benthic Diatom Samples. *Water Res.* **2001**, *35*, 2784–2788. [CrossRef]
61. Soininen, J. A Quantitative Analysis of Species Sorting across Organisms and Ecosystems. *Ecology* **2014**, *95*, 3284–3292. [CrossRef]
62. Epele, L.B.; Brand, C.; Miserendino, M.L. Ecological Drivers of Alpha and Beta Diversity of Freshwater Invertebrates in Arid and Semiarid Patagonia (Argentina). *Sci. Total Environ.* **2019**, *678*, 62–73. [CrossRef]
63. Bennion, H.; Sayer, C.D.; Tibby, J.; Carrick, H.J. Diatoms as Indicators of Environmental Change in Shallow Lakes. In *The Diatoms: Applications for the Environmental and Earth Sciences*; Cambridge University Press: Cambridge, UK, 2010.
64. Prygiel, J.; Carpentier, P.; Almeida, S.; Coste, M.; Druart, J.-C.; Ector, L.; Guillard, D.; Honoré, M.-A.; Iserentant, R.; Ledeganck, P.; et al. Determination of the biological diatom index (IBD NF T 90–354): Results of an intercomparison exercise. *J. Appl. Phycol.* **2002**, *14*, 27–39. [CrossRef]
65. Marzin, A.; Archaimbault, V.; Belliard, J.; Chauvin, C.; Delmas, F.; Pont, D. Ecological Assessment of Running Waters: Do Macrophytes, Macroinvertebrates, Diatoms and Fish Show Similar Responses to Human Pressures? *Ecol. Indic.* **2012**, *23*, 56–65. [CrossRef]
66. Blanco, S.L.; Ector, L.; Bécares, E. Epiphytic Diatoms as Water Quality Indicators in Spanish Shallow Lakes. *Vie Milieu* **2004**, *54*, 71–80.
67. Szczepocka, E.; Żelazna-Wieczorek, J.; Nowicka-Krawczyk, P. Critical Approach to Diatom-Based Bioassessment of the Regulated Sections of Urban Flowing Water Ecosystems. *Ecol. Indic.* **2019**, *104*, 259–267. [CrossRef]
68. Cantonati, M.; Angeli, N.; Virtanen, L.; Wojtal, A.Z.; Gabrieli, J.; Falasco, E.; Lavoie, I.; Morin, S.; Marchetto, A.; Fortin, C.; et al. Achnanthidium Minutissimum (Bacillariophyta) Valve Deformities as Indicators of Metal Enrichment in Diverse Widely-Distributed Freshwater Habitats. *Sci. Total Environ.* **2014**, *475*, 201–215. [CrossRef] [PubMed]
69. Kelly, M. The Semiotics of Slime: Visual Representation of Phytobenthos as an Aid to Understanding Ecological Status. *Freshw. Sci.* **2012**, *5*, 105–119. [CrossRef]
70. Eliasz-Kowalska, M.; Wojtal, A.Z. Limnological Characteristics and Diatom Dominants in Lakes of Northeastern Poland. *Diversity* **2020**, *12*, 374. [CrossRef]
71. Stenger-Kovács, C.; Hajnal, É.; Lengyel, E.; Buczkó, K.; Padisák, J. A Test of Traditional Diversity Measures and Taxonomic Distinctness Indices on Benthic Diatoms of Soda Pans in the Carpathian Basin. *Ecol. Indic.* **2016**, *64*, 1–8. [CrossRef]
72. Besse-Lototskaya, A.; Verdonschot, P.F.M.; Coste, M.; Van de Vijver, B. Evaluation of European Diatom Trophic Indices. *Ecol. Indic.* **2011**, *11*, 456–467. [CrossRef]
73. Poulíčková, A.; Letáková, M.; Hašler, P.; Cox, E.; Duchoslav, M. Species Complexes within Epiphytic Diatoms and Their Relevance for the Bioindication of Trophic Status. *Sci. Total Environ.* **2017**, *599–600*, 820–833. [CrossRef]
74. Poikane, S.; Zohary, T.; Cantonati, M. Assessing the Ecological Effects of Hydromorphological Pressures on European Lakes. *Inland Waters* **2020**, *10*, 241–255. [CrossRef]
75. Kelly, M. Data Rich, Information Poor? Phytobenthos Assessment and the Water Framework Directive. *Eur. J. Phycol.* **2013**, *48*, 437–450. [CrossRef]
76. Krztoń, W.; Kosiba, J. Variations in Zooplankton Functional Groups Density in Freshwater Ecosystems Exposed to Cyanobacterial Blooms. *Sci. Total Environ.* **2020**, *730*, 139044. [CrossRef] [PubMed]

ARTYKUŁ 3:

Eliasz-Kowalska, M., Wojtal, A. Z., Kryvosheia-Zacharova O. The epiphytic diatoms of the genus Gomphonema in Wigry National Park lakes (north-eastern Poland) (maszynopis)

The epiphytic diatoms of the genus *Gomphonema* in Wigry National Park lakes (north-eastern Poland)

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Abstract

Diatoms are increasingly used to assess water quality, as they are widely known as excellent bioindicators. They respond rapidly to changes in water physical and chemical characteristics and are sensitive to subtle changes in environmental conditions or disturbances that may affect other communities only at greater levels of disturbance. Knowledge of diatom taxonomy is a basic element of water quality evaluation. We analyzed the occurrence and abundance of *Gomphonema* taxa in ten selected characteristic lakes of Wigry National Park. The water lakes conditions there are very varied due to the location, history of shaping individual lakes, anthropogenic changes and water condition. The diversity of environmental conditions in lakes can be seen in the species composition of many diatom genera. However, the genus *Gomphonema* is one of the most diverse and well represented in all the lakes.

A total of 40 *Gomphonema* taxa were identified. Three of them were previously reported in Wigry National Park: *G. acuminatum*, *G. pumilum* and *G. vibrio*. Three of them are red-listed in Poland (*G. hebridense, G. pseudotenellum, G. vibrio*) and several taxa are poorly known from Europe (e.g. *G. angusticephalum*, *G. bavaricum, G. calcifugum, G. designatum, G. elegantissimum, G. geissleriae, G. graciledictum, G. insigniforme, G. jadwigiae, G. linearioides, G. qii, G. supertergestinum*). Some species showed a broad range of morphological variability, sometimes overlapping with similar taxa (e.g. *G. vibrio* group). The most *Gomphonema* taxa were present only in a few cells (e.g. *G. angusticephalum*, *G. designatum, G. geissleriae, G. graciledictum,* ***G. insigniforme***, *G. jadwigiae, G. qii*).

However, their presence enlarge our knowledge about their autecology and distribution pattern.

Introduction

The genus *Gomphonema* was erected by Ehrenberg in 1832. The genus is large, including over 500 taxa worldwide (Kociolek et al. 2019). The taxonomy of this genus can be difficult due to the phenotypic plasticity within species. However, some structural features e.g. raphe ultrastructure and their habitat preferences e.g. pH are useful in species identification or species complexes. The members of *Gomphonema* are an important component of the freshwater periphyton (Wojtal 2003, Novais 2009, Levkov et al. 2016, Noga et al. 2016, Bahls et al. 2018). Rapid progress in the genus morphotaxonomy has yielded descriptions of several new species (Reichard & Lange-Bertalot 1991, Reichardt 2015a, b, Levkov et al. 2016), part of which originated from species complexes ("catch-all taxa") (Reichard & Lange-Bertalot 1991, Reichardt 2015a, Levkov et al. 2016).

Some *Gomphonema* appear to be cosmopolitan, but some of them only occur in certain areas of the world. Depending on the species, they can occur in a wide range of environmental conditions, but many of them have clearly determined environmental tolerance ranges. Diatom assemblages are very sensitive to water chemistry; this makes them useful as indicators of environmental conditions (Resende 2005, Wojtal 2013). The species occurrence is also associated with trophic water level (oligotrophic, eutrophic or even polytrophic state) (Hofmann et al. 2011, Karthick 2011). So, diatoms are used for bioindication of specific environmental conditions (Hofmann et al. 2011, Bąk et al. 2012, Lange-Bertalot et al. 2017). Water quality assessments employ specific metrics, named - diatom indices. These metrics need to be based on autecological studies (Urrea & Sabater 2009, Lange-Bertalot et al. 2017). Estimation of the *Gomphonema* species diversity allows us to measure the differences among diatom assemblages present in every place, taking into account the identities of all the species.

Wigry National Park (WPN) is named after lake Wigry, the largest of the Park's many lakes. Wigry Lake is one of the earliest places studied on a broad limnological scale in Poland. The establishment of the Warsaw Scientific Society's hydrobiological station at Płociczno in 1920 led to hundreds of publications. There are only a few papers about the diatoms of Wigry Lake. The most detailed data are in the oldest works (e.g., Wołoszynska 1922, 1923; Wisłouch 1926). Researches were done on recent benthic diatoms (Wołoszynska 1923), phytoplankton

(Wołoszynska 1922; Wisłouch 1926) and lake sediments (Witkowski 2009). All together, about 200 diatom taxa have been identified from Wigry lake, including seven *Gomphonema* taxa: *G. insigne, G. pumilum* and *G. vibrio* (Wołoszyńska 1922) and *G. acuminatum, G. angustatum, G. constrictum*, *G. gracile* (Wołoszyńska 1923; Tomaszewicz 1996). Amongst them only two species were found (*G. pumilum* and *G. vibrio*) during the current research on ten very different lakes of the WPN. The variability of diatom assemblages is additionally related to the seasons of the year (Elias et al. 2012, Hassan 2018). This variability is not always related to the availability of nutrients. Hydrological regime, light, temperature and grazing are also affecting phytobenthos (Kelly et al. 2009, Rimet et al. 2015).

The aim of present study was to determine the diversity of the epiphytic genus *Gomphonema* in Wigry National Park lakes. The sampling sites were distributed over the whole Park area, and differed in water chemistry. Diatoms of the genus *Gomphonema* were found in every sample collected during this study.

Study area

The Wigry National Park is located close to the Lithuanian border, in the Suwalki Lakeland (54,09°N, 23,03°E) at 131.9 m above sea level (Fig. 1). The length of the growing season for the Suwalki region is more than a month shorter than in other parts of Poland. The Wigry National Park covers parts of the Masurian Lake District and Augustów Primeval Forest (Puszcza Augustowska). The study area is under influence of a temperate climate transitional between the maritime and the continental (Andrzejczyk and Brzeziecki, 1995; Drzymulska and Zieliński, 2014). Studied lakes are of glacial and postglacial origin and are remnants of Weichselian Glaciation (Górniak, 2006). Despite lakes differentiate in limnological, physical and chemical features, they are all characterized by harmonic evolution.

Trophic state of Białe Pierciańskie lake and Okrągłe lake was estimated as eutrophic whereas Suchar Wielki lake, Suchar III lake, Wądołek lake and Wygorzele lake, as dystrophic. The last four lakes are peat bog lakes and differ in water parameters (Fig. 1, Tab. 1). The biggest lake is Wigry – 2163.3ha and the smallest is Okrągłe lake – 13.7ha (Table 1). Direct catchment of the lakes also differentiate. The Okrągłe Lake and Wigry Lake are more affected by human impact, whereas Krusznik Lake is impacted by extensive agriculture. Wigry lake was formed during the Baltic phase of the last glaciation; this accounts for its varied morphometry. The Czarna Hańcza river, Wiatrołuża river and other streams flow into the Wigry lake, and the Czarna Hancza drains it (Bajkiewicz-Grabowska 2009). Habitat conditions vary due to spatial differentiation of the lake's shape, trophic state, thermal and oxygen conditions, and water exchange (Migaszewski 2003). Among the most important dominant macrophytes are common *Phragmites australis* Trin. ex Steud. (= *Phragmites communis* Trin.) and *Carex* ssp.

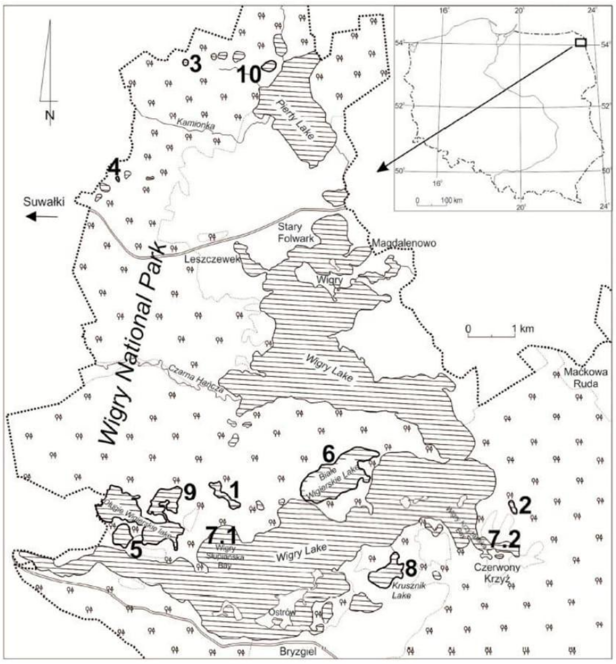


Figure 1. Distribution of investigated localities: 1 - Suchar Wielki (SW), 2 - Wygorzele (WYG), 3 - Wądołek (WAD), 4 - Suchar III (SIII), 5 - Okrągłe (OK), 6 - Białe Wigierskie (BW), 7 - Wigry (7.1 - Słupiańska Bay (WS), 7.2 - Krzyżańska Bay (WK), 8 - Krusznik (K), 9 - Muliczne (M), 10 - Białe Pierciańskie (BP) lakes.

Lake	Area [ha]	Max. depth [m]	Coastline [m]	Direct catchment [ha]	Catchment [ha]
Wigry	2163.3	73	63920	5159.8	45293.1
Krusznik	26.7	18	2643	70.7	70.7
Białe Wigierskie	99.9	34	5117	329.1	329.1
Muliczne	24.1	11.3	3175	191.2	191.2
Okrągłe	13.7	13	1459	28.5	906.8
Białe Pierciańskie	6.9	24	1011	50.4	50.4

Wygorzele	2	3	670	63.5	63.5
Suchar Wielki	8.44	9.6	2066	107.1	107.1
Wądołek	1.09	15	474	19.4	19.4
Suchar III	0.44	4	320	32.2	32.2

Table 1. Description of the physical characteristics of the studied lakes.

Lake	pH	Conductivity	Cl^-	CO_3^{2-}	SO_4^{2-}	NO_3^-	NH_4^+	Mg^{2+}	PO_4^3	Ca^{2+}
		µS/cm	[mg/L]							
O	7.68–8.28	338–385	8.45–9.99	103.52–248	12.21–30.92	0.48–1.75	0.04–0.83	11.58–15.58	0.002–0.02	46.81–73.62
M	7.52–8.26	294–342	3.52–3.85	126.70–239	21.35–24.06	0–0.37	0.02–0.22	10.14–14.45	0.002–0.011	40.21–68.25
BP	7.63–8.23	385–401	2.28–2.94	186.77–292	3.15–5.63	0–0.35	0.02–0.32	13.88–18.95	0.002–0.01	48.46–69.45
BW	7.32–8.15	164–185	2.68–3.35	74.81–137.4	6.35–6.81	0.00–0.36	0.01–0.13	4.93–6.53	0.002–0.03	23.85–36.66
K	7.51–8.26	234–342	4.24–4.52	101.49–239	12.36–22.23	0–0.35	0.036–0.23	7.66–14.45	0.002–0.01	30.45–68.25
W	7.56–8.23	350–402	14.47–17.05	135.36–243	21.68–23.57	0–1.26	0.012–0.12	11.80–16.08	0.002–0.03	16.05–45.67
WYG	3.7–6.11	20–25	1.40–2.03	8.44–35.71	0.02–1.23	0.00–0.40	0.20–1.26	0.00–0.49	0.01–0.03	0.85–2.01
WAD	4.65–5.31	20–23	1.27–1.35	17.56–18.60	0.02–0.56	0–0	0.00–0.20	0.77	0.005–0.04	2.67–3.35
SIII	3.6–6.8	22–23	0.86–8.44	9.04–18.45	0.02–1.11	0–0	0.08–0.20	0.00–0.46	0.02–0.05	0.59–1.74
SW	4.60–6.34	15–18	0.97–1.28	8.66–39.66	2.23–2.68	0.00–0.36	0.00–0.00	0.50–0.74	0.01–0.02	1.73–2.75

Table 1. Chemical water parameters of the ten lakes (Wigierski National Park): O – Okrągłe Lake, M – Muliczne Lake, BP – Białe Pierciańskie Lake, BW – Białe Wigierskie Lake, K – Krusznik Lake, W – Wigry Lake, WYG – Wygorzele Lake, WAD – Wądołek Lake, SIII – Suchar III Lake, SW – Suchar Wielki Lake.

Materials and methods

A total of 92 water samples were analyzed for their physical and chemical properties. They were collected in spring (May 2015, May 2017, May 2018), summer (September 2017, October 2017) and fall (November 2016, September 2018) from 11 sampling points (in each lake, in Wigry lake two localities). Collection sites were located in: Suchar Wielki (SW), Wygorzele (WYG), Wądołek (WAD), Suchar III (SIII), Okrągłe (OK), Białe Wigierskie (BW), Wigry [Słupiańska Bay (WS), Krzyżańska Bay (WK)], Krusznik (K), Muliczne (M) and Białe Pierciańskie (BP) (Figure1). All sampling for physical and chemical properties were done in open water, with comparable environments selected as far as possible. Conductivity and pH were measured using a YSI 6600 V2 multiparameter sonde. Water for chemical analyses was sampled 20-30cm below the lake surface using 0.33 L polyethylene bottles, and stored in the dark at 4°C, to limit ongoing chemical reactions. Ionic analyses were related to PO_4^{3-}, SO_4^{2-}, NO_3^{-}, F^{-}, CO_3^{2-}, Cl^{-} and NO_2^{-}, as well as Na^{+}, Li^{+}, K^{+}, Mg^{2+}, NH_4^{+} and Ca^{2+} (Table 2.) Laboratory measurement involved the *Dionex* ion chromatograph at the laboratory of the Institute of Nature Conservation, Polish Academy of Sciences.

The diatom material was taken at the same time as the water samples. The material was collected from the macrophytes *Phragmites australis* (Cav.) Trin. ex Steud. (Okrągłe, Białe Wigierskie, Wigry, Krusznik, Muliczne and Białe Pierciańskie lakes) and *Carex* spp. (Suchar III, Suchar Wielki, Wądołek and Wygorzele lakes). Samples were cleaned by addition of 37% H_2O_2, and then heated. The cleaning was completed by adding $KMnO_4$ and HCl. The cleaned diatom material dried on slides was mounted in Naphrax® synthetic resin. The slides were then analysed using an Nikon Eclipse-80i microscope, with 400 diatom valves counted. Diatom identification mainly followed Hofmann et al. (2011), Wojtal et al. (2011), Bąk et al. (2012), Lange-Bertalot and Ulrich (2014), Van De Vijver (2014), Delgado et al. (2015), Lange-Bertalot et al. (2017), Kennedy and Allott (2017) and Heudre et al. (2019).

Results

The epiphytic genus *Gomphonema* in Wigry National Park lakes was very rich in several rare taxa. The sampling sites were distributed over the whole Park area, and differed in water chemistry. Most of them (Table 3) was found in the Wigry lake, what is related to its large area. In this study, 40 taxa of the genus *Gomphonema* were identified. Three of the taxa are red-listed in Poland (*G. hebridense, G. pseudotenellum, G. vibrio*). Most of the taxa belong, however, to very rarely reported diatoms.

Gomphonema acuminatum Ehrenberg 1836 var. ***acuminatum***

Refs Hofmann et al. (2011), p. 294, plate 93: 9-12; Bąk et al. (2012) p. 165, plate 64, Krammer & Lange-Bertalot (1986) p. 365, plate 160: 1, 2.

The valves are 29-49 µm long and 7-10 µm wide, clavate and tumid at the mid-valvae with two constrictions along the margins. The striae are radial throughout the valve, becoming more radial towards the poles, 10-11 in 10 µm. The raphe is lateral, undulate and runs in narrow, straight axial area.

Gomphonema acuminatum is known from Wigry lake (Wołoszyńska 1924; Tomaszewicz 1996 and outflow of the Lake Okrągłe (Plan Ochrony dla Wigierskiego Parku Narodowego 2011). The species is known from many other localities in Poland, where water was of oligo-mesosaprobic and oligo- mesoeutrophic order, of moderate water specific conductivity.

Gomphonema angustum C. Agardh 1831

Refs Hofmann et al. (2011), p. 296, figs 97 (23-27); Bąk et al. (2012) p. 166, figs 67, Krammer & Lange-Bertalot (1986) p. 370, figs 164 (1, 2).

Valves are 23-26 µm long and 5.3-6.0 µm wide, with rounded apices. The striae are located more dense near the poles and more widely towards the valve mid-portion, slightly radiate, 9-12 in 10 µm. Central area is large, with one stigma.

Gomphonema angustum was found in Wigry lake in May, where its relative abundance reached 1.8% (uncommon). This taxon has been not yet reported from the Wigry Lake though is known from the outflow of tha Lake Okrągłe (Plan Ochrony dla Wigierskiego Parku Narodowego 2011). The species is an alkalophilous, oligotrophic and oligo- mesosaprobous diatom and is quite common in Poland in waters rich in calcium.

Gomphonema auritum A. Braun ex Kützing 1849

Refs Hofmann et al. (2011), p. 297, figs 98 (16-20); Bąk et al. (2012) p. 167, figs 67, Krammer & Lange-Bertalot (1986) p. 358, figs 154 (26,27).

Valves are lanceolate, clavate, 34–43 µm long and 5.7–6.0 µm wide. The striae become more radiate towards the upper pole and less radiate towards the lower pole, 11-12 in 10µm.

During the study *G. auritum* was found in the Bryzgiel side, in May, its abundance reached 1% (uncommon). This taxon has been not reported previously from Wigry Lake. The distribution of *G. auritum* remains uncertain as it can be misidentified with *G. gracile*.

However, the current data show *G. auritum* preference of lakes rich in calcium with oligo-mesotrophic waters, mainly in mountain areas.

Gomphonema brebissonii Kützing 1849

Refs Hofmann et al. (2011), p. 298, plate 93: 16-18; Bąk et al. (2012) p. 168, plate 64, Krammer & Lange-Bertalot (1986) p. 365, Plate 160: 3, 4.

Valves are clavate, slightly constricted above the middle 40–80 µm long and 7.8–11.0 µm wide. The striae are ornamented parallel in the mid-valve becoming to radial towards the poles 10–11 in 10 µm.

Gomphonema brebissonii was recorded for the first time for Wigry lake. The species is known from oligosaprobic and oligo- mesosaprobous waters in Poland (Wojtal 2003), in low-moderate values of specific water conductivity.

Gomphonema calcareum P.T. Cleve 1868

Refs Hofmann et al. (2011), p. 309, figs 95 (15,16); Bąk et al. (2012) p. 177, figs 65, Krammer & Lange-Bertalot (1986) p. 374, figs 165 (9, 10).

The valves are lanceolate-clavate, 42-44 µm long and 8.0-9.0 µm wide; with a broadly rounded headpole and footpole. The axial area is narrow and linear. The central area is rather large, abruptly formed by shortened bilaterally striae, without any stigma. The transapical striae are radial becoming subparallel towards the headpole, 10 in 10 µm.

Gomphonema calcareum was found as an uncommon diatom. This species was found for the first time in Wigry lake.

Gomphonema calcifugum Lange-Bertalot & Reichardt in Lange-Bertalot & Genkal 1999

Refs Hofmann et al. (2011), p. 299, figs 96 (33-37); Bąk et al. (2012) p. 168, figs 66.

Valves are clavate with a broadly rounded headpole, 18-29 µm long and 4.0-6.5 µm wide. The axial area is narrow and straight, forming a rectangular central area bordered by shortened striae at or near the margin. The striae are radiate, more strongly near the mid-valve portion, 11-13 in 10 µm. Part of the observed valves were narrower (since 4 µm) than is provided in Krammer and Lange-Bertalot (1986).

The species has been not yet reported from Wigry lake.

Gomphonema hebridense Gregory 1854

Refs Hofmann et al. (2011), p. 303, figs 98 (6-10); Bąk et al. (2012) p. 172, figs 67, Krammer & Lange-Bertalot (1986) p. 362, figs 156 (12, 13).

Valves are linear-lanceolate and clavate, 22-45 µm long and 4.9-6.0 µm wide. The striae are parallel near the valve poles and very weakly radiate towards the mid-valve portion; 13-17 in 10 µm. Axial area is narrow and linear. Central area is formed by unilaterally shortened stria. This is the first report of this species in Wigry Lake. The distribution in Poland remains to be checked. This taxon is noted in Red list of plants and fungi in Poland as Rare (R).

Gomphonema lateripunctatum Reichardt & Lange-Bertalot 1991

Refs Hofmann et al. (2011), p. 305, figs 95 (25-30); Bąk et al. (2012) p. 173, figs 66.

Valves are linear-lanceolate to clavate; 30-45 µm long and 6 µm broad. Striae are radiate at the mid-valve portion, becoming more strongly radiate and closer distributed at the headpole and footpole; 10-11 in 10 µm. The axial area is of a various size, lanceolate. The central area is slightly asymmetric and smaller on the side bearing the stigma.

This is the first report of the species in the Wigry Lake. *Gomphonema lateripunctatum* is a common diatom in rich in calcium, oligo- mesotrophic waters, being the taxon that indicates a very good ecological value of inhabited environments.

Notes: Some differences in the dimensions of this taxon were found, eg. the observed valves were narrower than *G. lateripunctatum*, which according to Reichardt and Lange-Bertalot (1991) should have 8-14 µm of the width.

Gomphonema minusculum Krasske 1932

Refs Hofmann et al. (2011), p. 306, figs 96 (16-21); Bąk et al. (2012) p. 175, figs 66,

Valves are cuneate and lanceolate, 19-23 µm long and 3.0-4.3 µm wide, with a weakly radiate striae throughout the whole valve 13-15 in 10µm.

Gomphonema minusculum was found for the first time for the Wigry Lake. This taxon inhabits environments of a broad ecological status, e.g. from oligo- up to eutrophic waters.

Gomphonema olivaceoides Hustedt 1950 Refs Hofmann et al. (2011), p. 310, figs 95 (10-14); Bąk et al. (2012) p. 177, plate 65, Krammer & Lange-Bertalot (1986) p. 375, figs 165 (14-18).

The valves are clavate with bluntly rounded headpole, 17-34 µm long and 5.5-6.0 µm wide The striae are weakly radial throughout the valvae 13-16 in 10 µm. Central area is rectangular, bordered by shortened striae at, or near, the valve margin.

Gomphonema olivaceoides has been not yet reported from the Wigry Lake.
Notes. Part of the observed specimens have a higher striae density than provided in the keys, ie. up to 16 striae in 10 µm.

Gomphonema pala Reichardt 2001

Refs Hofmann et al. (2011), p. 305, figs 94 (16-20); Bąk et al. (2012) p. 178, figs 65.
The valves are strongly clavate, with broadly rounded, distinct headpole and distinctively broadened mid-valve portion, 26-36 µm long and 9-11 µm wide. The striae are radiate throughout the whole valve, 12-13 in 10 µm, near the footpole more radiate and more densely located. The axial area is linear, with a lateral raphe branches. Central area is clearly visible, with stigma and raphe branches ends.
Gomphonema pala is widespread in mountain lakes and similar environments, though it has been not reported previously from Wigry lake.

Gomphonema parvulum var. ***parvulum*** (Kützing) Kützing 1849

Refs Hofmann et al. (2011), p. 312, figs 99 (1-5); Bąk et al. (2012) p. 179, figs 68, Krammer & Lange-Bertalot (1986) p. 358, figs 154 (1, 2).
Valves are clavate and subrostrate, 21-25 µm long and 5-6 µm wide. The striae are weakly radiate throughout a valve, up to 12 in 10 µm. Axial area is narrow and linear. One shortened striae form transapical rectangular central area with distinct punctum.
This taxon has been not yet reported from the Wigry Lake though belongs to the group of the most common diatoms of the streams of Wigierska refuge (Maniówka, Gremzdówka, Czarna Hańcza in Sobolewo, Czarna Hańcza outflow and Czarna Hańcza – Bindużka) (Plan Ochrony dla Wigierskiego Parku Narodowego 2011). *Gomphonema parvulum* is known from oligosaprobic and mesosaprobic waters of various trophic state (e.g. Bąk et al. 2012).

Gomphonema procerum Reichardt & Lange-Bertalot 1991

Refs Hofmann et al. (2011), p. 313, figs 96 (27); Bąk et al. (2012) p. 179, plate 66.
The valves are narrowly lanceolate, slightly broaded in the mid-valve portion, 38-57 µm long and 5.7-6.6 µm wide. The footpole is of ca. half wideness of the headpole. The transapical striae are a very slightly radiate, becoming (sub-)parallel in the mid-valve portion, 11-12 in 10 µm. Axial area is narrow and linear. Central area is small but distinct.
Gomphonema procerum has been not reported from the Wigry lake so far.

Gomphonema pseudotenellum Lange-Bertalot in Krammer & Lange-Bertalot 1985

Refs Hofmann et al. (2011), p. 302, figs 99 (25-29); Bąk et al. (2012) p. 180, figs 66, Krammer & Lange-Bertalot (1986) p. 372, figs 164 (22-24).

The valves are narrowly lanceolate, with a narrow axial area and small central area. The only one valve found was 23 long and 3.2 wide with 12.5 striae in 10 µm. *Gomphonema pseudotenellum* was identified as the red-listed in Poland, where this species is classified as endangered. This diatom was found as a rare species and has been not reported from Wigry lake, so far.

Gomphonema supertergestinum Reichardt 2009

Refs Hofmann et al. (2011), p. 318, figs 96 (22-26); Bąk et al. (2012) p. 182, figs 66, Krammer & Lange-Bertalot (1986) p. 373, figs 162 (6,7).

The valves are lanceolate-clavate, 40-47µm long and 8.0-9.5 µm wide; with a broady rounded headpole and footpole with pseudoseptum. The axial area is narrow and linear. The central area is large, abruptly formed, with distinct stigma located between central raphe ends, and shortened striae unilaterally formed. The transapical striae are radial becoming subparallel towards the headpole, 11-12 in 10 µm.

Gomphonema supertergestinum was not recorded previously from Wigry lake.

Gomphonema vibrio Ehrenberg 1843

Refs Hofmann et al. (2011), p. 319, figs 96 (1-5); Bąk et al. (2012) p. 184, figs 66.

The large valves, 40-97 µm long and 7-10 µm wide with 7-11 striae in 10 µm. The striae are weakly radial in the mid-valve portion and becoming more radiate towards the headpole and less radiate (to parallel) near a footpole. Raphe branches are lateral with a proximal raphe branches located near the central area border.

Gomphonema vibrio was found in the Słupiańska bay, Bryzgiel, Klasztor, Wigry Wieś (0,74% - rare) and Uklei bay. Previously reported by Wołoszyńska (1922) and Tomaszewicz (1996) from Wigry lake. This taxon is listed in the Red list of plants and fungi in Poland as Endangered (E).

Gomphonema vibrio inhabits oligo- mesotrophic lake waters, which are rich on calcium ions. This taxon is known as a very good bioindicator of ecological status of inhabited waters.

Table 3. Occurrence of *Gomphonema* taxa recorded at 10 lakes. 1 - Suchar Wielki (SW), 2 - Wygorzele (WYG), 3 - Wądołek (WAD), 4 - Suchar III (SIII), 5 - Okrągłe (OK), 6 - Białe Wigierskie (BW), 7 - Wigry (W), 8 - Krusznik (K), 9 - Muliczne (M), 10 - Białe Pierciańskie (BP) lakes.

Plate 1. Fig. 2-42, 2,3 – *Gomphonema acuminatum* Ehrenberg; 4,5 – *G. angusticephalum* Reichardt & Lange-Bertalot; 6,7 – *G. angustum* Agardh; 8,9 – *G. auritum* A.Braun ex Kützing; 10-13 – *G. bavaricum* Reichardt & Lange-Bertalot; 14,15 – *G. brebissonii* Kützing; 16-18 – *G. calcareum* P.T. Cleve; 19-22 – *G. calcifugum* Lange-Bertalot & Reichardt; 23,24 – *G. capitatum* Ehreberg; 25,26 –*G. clavatulum* Reichardt; 27 – *G.* cf. *clavatum* Ehrenberg; 28,29 – *G. designatum* Reichardt; 30 – *G. dichotomum* Kützing; 31,32 – *G. elegantissimum* Reichardt & Lange-Bertalot; 33-35 – *G. exilissimum* (Grunow) Lange-Bertalot & Reichardt; 36,37 – *G. geissleriae* Reichardt & Lange-Bertalot; 38-42 – *G. graciledictum* Reichardt; 1-41 LM; 42 SEM. Scale bars = 10 µm.

Plate 2. Fig. 43-60, 43-45 – *G. hebridense* Gregory; 46 – *G. insigniforme* Reichardt & Lange-Bertalot; 47-49 – *G. italicum* Ehrenberg; 50-52 – *G. jadwigiae* Lange-Bertalot & Reichardt; 53-56 – *G. lateripunctatum* Reichardt & Lange-Bertalot; 57-60 – *G. minusculum* Krasske; 43-51, 53, 54, 57-60 LM; 52, 55, 56 SEM. Scale bars = 10 µm.

Plate 3. Fig. 61-77, 61-62 – *G. linearioides* Levkov (= *Gomphonella*); 63-64 – *G. minutum* (Agardh) Agardh; 65-69 – *G. olivaceoides* (Hustedt) Lange-Bertalot; 70-74 – *G. olivaceum* (Hornemann) Brébisson; 75-77 – *G. parvulum* (Kützing) Kützing; 61-67, 70-72, 75-77 LM; 68, 69, 73, 74 SEM. Scale bars = 10 µm.

Plate 4. Fig. 78-94, 78-80 – *G. pala* Reichardt; 81-83 – *G. pratense* Lange-Bertalot & Reichardt; 84-87 – *G. procerum* Reichardt & Lange-Bertalot; 88 – *G. pseudopusillum* Reichardt; 89 – *G. pseudotenellum* Lange-Bertalot; 90-94 – *G. pumilum* (Grunow) Reichardt & Lange-Bertalot; 78-82, 84-92 LM; 83, 93, 94 SEM. Scale bars = 10 µm.

Plate 5. Fig. 95-108, 95-96 – *G. qii* Juttner; 97-99 – *G. subangustatum* Lange-Bertalot, Cavacini, Tagliaventi & Alfinito; 100 – *G.* cf. *subclavatum* (Grunow) Grunow; 101,102 – *G. supertergestinum* Reichardt; 103 – *G. vibrio* var. *subcapitatum* (Mayer) Lee; 104,106 – *G. vibrio* Ehrenberg; 107,108 – *G. dichotomum* Kützing; 95-107 LM; 108 SEM. Scale bars = 10 µm.

Disscussion

Diatoms are increasingly used to assess water quality, as they respond rapidly to changes in water physicochemistry and are sensitive to subtle changes in environmental conditions or disturbances that may affect other communities only at greater levels of disturbance (e.g., Dixit et al. 1992). A diatom community's species composition and abundance are likely to be altered by changes in water properties (Clements 1991). Diatoms are excellent biological indicators for many types of pollution in aquatic systems, such as heavy metal contamination (Patrick & Palavage 1994; Kelly et al. 1995). High growth rates in the periphyton allow communities to be completely replaced in a few weeks in response to environmental factors, so they reflect recent water conditions (Rott, 1991; Round, 1991). Knowledge of the structure of diatom assemblages is a key element of water quality assessment. We analyzed the occurence and abundance of *Gomphonema* taxa in Wigry Lake, as they occurred abundantly in the analyzed samples. We identified 19 *Gomphonema* taxa, three of which were previously reported in Wigry National Park: *G. acuminatum* (Woloszynska 1923; Tomaszewicz 1996), *G. pumilum* (Woloszynska 1922) and *G. vibrio* (Woloszynska 1922). Three of the identified species are red-listed in Poland, where they are classified as endangered (*Gomphonema pseudotenellum, G. vibrio*) and rare (*G. hebridense*) (Siemińska 2006). The rare diatom *G. hebridense* is not endangered or vulnerable at this time but is at risk of becoming so. One taxon was very common (*G. pumilum*), eleven were uncommon (*G. angustum, G. auritum, G. brebissonii, G. calcareum, G. calcifugum, G. hebridense, G. lateripunctatum, G minusculum, G. pala, G. parvulum* var. *parvulum, G. superteregestinum*), and six were rare (*G. acuminatum, G. olivaceoides, G. procerum, G. pseudotenellum, G. vibrio*). *Gomphonema pumilum* was in the samples from almost every sampling site. We did not find four of seven *Gomphonema* taxa previously reported from Wigry National Park: *G. angustatum, G. constrictum*, *G. insigne* and *G. gracile*. Some taxa new for Wigry Lake were identified, among which were the recently described *G. supertergestinum* (Reichardt 2009) and the recently revised *G. calcareum* (Reichardt, 2009), *G. calcifugum* (Reichardt, 2009), *G. olivaceoides* (Reichardt, 2009) and *G. pumilum* (Reichardt, 2009b). The recent investigation of *G. acuminatum* species complex from the Lake Glubokoe revealed presence of unknown *G. megalobrebissonii* and a few unidentified taxa (Chudaev et al. 2014). *Gomphonema parvulum* is worldwide taxon which need to be revised because it probably represent morphologically similar but separate taxa. *Gomphonema lateripunctatum* also requires revision because the observed valves were narrower than the width values ranging from 8 µm to

14 µm given by Reichardt and Lange-Bertalot 1991. This difference can be seen in published photos from other studies (Bąk et al., 2012; Hofmann et al., 2011).

Conclusions

1. In this study, 40 taxa of the genus *Gomphonema* were identified in samples from Wigry National Park lakes.

2. Three of those taxa were previously reported in Wigry National Park (*G. acuminatum, G. pumilum, G. vibrio*) (Wołoszynska 1922, 1923, 1925).

3. Three of the taxa are red-listed in Poland (*G. hebridense, G. pseudotenellum, G. vibrio*). (Siemińska et al. 2006). Most of the taxa belong, however, to very rarely reported diatoms.

4. The observed taxa can found in calcium-rich meso-eutrophic lakes such as Wigry Lake.

References

1. Dixit S, Smol J, Kingston J, Charles D. Diatoms: powerful indicators of environmental change. Environ Sci Technol. 1992;26:21–33. http://dx.DOI.org/10.1021/es00025a002
2. Wołoszyńska J. Zimowa flora Wigierskich źródeł morenowych. Kosmos. 1922;47(1–3):305–326.
3. Wołoszyńska J. Rozmieszczenie glonów osiadłych na dnie jeziora Wigierskiego. Część I. Spraw. Stacji Hydrobiol. Na Wigrach. 1923;1(2–3):2–66.
4. Wołoszyńska J. Notatki algologiczne. Spraw. Stacji Hydrobiol. na Wigrach. 1925;1(4):3–9.
5. Zarzycki K. Mirek Z. Red list of plants and fungi in Poland. Czerwona lista roślin i grzybów Polski. Kraków: Instytut Botaniki im. W. Szafera PAN; 2006.
6. Resende P, Azeiteiro U, Pereira MJ. Diatom ecological preferences in shallow temperate estuary (Ria de Aveiro, Western Portugal). *Hydrobiologia*. 2005;*544:77–88.* DOI: 10.1007/s10750-004-8335-9
7. Urrea G, Sabater S. Epilithic diatom assemblages and their relationship to environmental characteristics in an agricultural watershed (Guadiana River, SW, Spain). Ecol. Indic. 2009;9:693–703. DOI: 10.1016/j.ecolind.2008.09.002
8. Müller, U. The vertical zonation of adpressed diatoms and other epiphytic algae on *Phragmites australis*. Eur. J. Phycol. 1999;34: 487–496.
9. Novais MH, Blanco S, Hlubikova D, Falasco E, Gomá J, Delgado C, et al. Morphological examination and biogeography of the *Gomphonema rosenstockianum* and *G. tergestinum* species complex (Bacillariophyceae). Fottea. 2009;9: 257–274. DOI: 10.5507/fot.2009.026

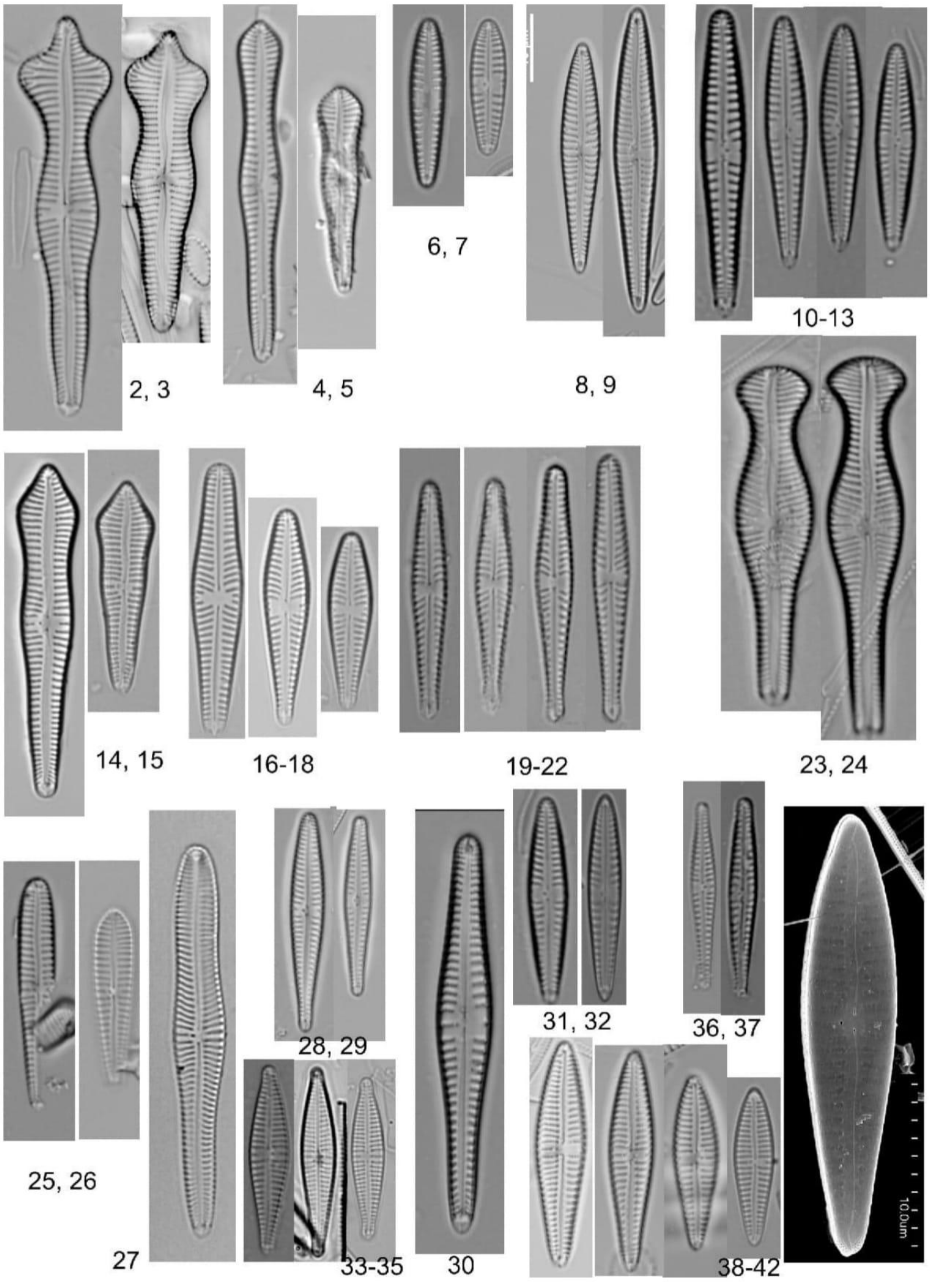
10. Karthick B, Kociolek JP, Mahesh MK, Ramachandra TV. The diatom genus *Gomphonema* in India: Checklist and description of three new species. Nova Hedwigia. 2011;93:235–264. http://dx.DOI.org/10.1127/0029-5035/2011/0093-0211
11. Witkowski A, Bąk M, Kierzek A. Ewolucja jeziora od schyłku ostatniego zlodowacenia : zmiany warunków środowiskowych w holocenie w Jeziorze Wigry w świetle analizy diatomologicznej osadów. In: Rutkowski J, Krzysztofiak L, editors. Jezioro Wigry : historia jeziora w świetle badań geologicznych i paleoekologicznych. Suwałki: „Man and Nature" Association; 2009. p. 215–226.
12. Wisłouch S. O letnim fitoplanktonie Jezior Wigierskich. Arch. Hydrobiol. Rybactwa. 1926;1(1–2):79–114.
13. Besse–Lototskaya A, Verdonschot PF, Coste M, Van de Vijver B. Evaluation of European diatom trophic indices. Ecol. Indic. 2011;11:456–467. DOI: 10.1016/j.ecolind.2010.06.017
14. Rutkowski J, Król K, Szczepańska J. Lithology of the profundal sediments in Słupiańska Bay (Wigry Lake, NE Poland) – introduction to interdisciplinary study. Geochronometria. 2007;27:47–52. DOI: 10.2478/v10003-007-0020-3
15. Piotrowska N, Hajdas I, Bonani G. Construction of the calendar time scale for Lake Wigry (NE Poland) sediments on the basis of radiocarbon dating. Radiocarbon. 2007;49(2):1133–1143.
16. Bajkiewicz–Grabowska E. Water circulation in a lake at extreme water levels: Lake Wigry case study, Limnol. Rev. 2009;9(2–3):63–72.
17. Migaszewski Z, Gałuszka A. Skład chemiczny wód i osadów z wybranych rzek z Wigierskiego Parku Narodowego i rejonu Suwałk oraz osadów Jeziora Wigry. 2003. Available from: http://www.wigry.org.pl/chem/index.htm
18. Górniak A, editor. Jeziora Wigierskiego Parku Narodowego. Aktualna jakość i trofia wód. Białystok: Wyd. Uniwersytetu w Białymstoku; 2006.
19. Picińska–Fałtynowicz J, Błachuta J. Wytyczne metodyczne do przeprowadzenia oceny stanu ekologicznego jednolitych części wód rzek i jezior oraz potencjału ekologicznego sztucznych i silnie zmienionych jednolitych części wód płynących Polski na podstawie badań fitobentosu. Zakład Ekologii; Instytut Meteorologii i Gospodarki Wodnej, Wrocław, 2010.
20. Krammer K, Lange–Bertalot H. Bacillariophyceae. 1. Teil: Naviculaceae. In Ettl H, Gerloff J, Heynig H, Mollenhauer D, editors. Süsswasser flora von Mitteleuropa, Band 2/1. Stuttgart, New York: Gustav Fischer Verlag; 1986.
21. Hofmann G, Lange–Bertalot H, Werum M. Diatomeen im Süβwasser–Benthos von Mitteleuropa. Königstein: A.R.G. Gantner Verlag K.G; 2011.
22. Bąk M, Witkowski A, Żelazna–Wieczorek J, Wojtal A, Szczepocka E, Szulc K, et al. Klucz do oznaczania okrzemek w fitobentosie dla potrzeb oceny stanu ekologicznego
23. wód powierzchniowych w Polsce. Szczecin: Główny Inspektorat Ochrony Środowiska; 2012.
24. Tomaszewicz G. Flora glonów Wigierskiego Parku Narodowego. Fragm. Flor. Geobot. Ser. Polonica. 1996;3:239–259.
25. Patrick R, Palavage DM. The value of species as indicators of water quality. Proceedings of the Academy of Natural Sciences of Philadelphia. 1994;145:55–92

26. Kelly MG, Penny CJ, Whitton BA. Comparative Performance of Benthic Diatom Indices Used to Assess River Water Quality. Hydrobiologia. 1995;302:179–188. DOI: 10.1007/BF00032108
27. Clements WH. Community responses of stream organisms to heavy metals: A review of observational and experimental approaches. In Newman MC, McIntosh AW editors.. Metal Ecotoxicology Concepts & Applications. Orlando: Lewis Publ; 1991. p. 363–391.
28. Rott E. Methodological aspects ands perspectives in the use of periphyton for monitoring and protecting rivers. In Whitton BA, Rott E, Friedrich G, editors. Use of Algae for Monitoring Rivers I. Düsseldorf, Germany; 1991. p. 9–16.
29. Round FE. Diatoms in river water–monitoring studies. J. appl. Ecol. 1991;3:129–145. DOI: 10.1007/BF00003695
30. Reichardt E. Silikatauswüchse an der inneren Stigmenöffnungen bei *Gomphonema*–Arten. Diatom Res. 2009;24(1):159–173. DOI: 10.1080/0269249X.2009.9705788
31. Passy SI, Kociolek JP, Lowe RL. Five new *Gomphonema* species (Bacillariophyceae) from rivers in South Africa and Swaziland. J Phycol. 1997;33: 455–474. DOI: 10.1111/j.0022-3646.1997.00455.x
32. Jüttner, I., Ector, L., Reichardt, E., van de Vijver, B., Jarlman, A., Krokowski, J. & Cox, E.J. *Gomphonema varioreduncum* sp. nov., a new species from northern and western Europe and a reexamination of *Gomphonema exilissimum*. Diatom Res. 2013;28:303–316. DOI: 10.1080/0269249X.2013.797924
33. Abarca N, Jahn R, Zimmermann J, Enke N. Does the Cosmopolitan Diatom *Gomphonema parvulum* (Kützing) Kützing Have a Biogeography? PloS ONE. 2014;9:e86885. DOI: 10.1371/journal.pone.0086885
34. Reichardt E. New and recently described *Gomphonema* species (Bacillariophyceae) from Siberia. Fottea. 2009;9(2):289–297. DOI: 10.5507/fot.2009.028

Fig.	Taxon	Lake									
		SW	WYG	WAD	SIII	OK	BW	W	K	M	BP
2, 3	*Gomphonema acuminatum* Ehrenberg							+			
4, 5	*G. angusticephalum* Reichardt & Lange-Bertalot			+							
6, 7	*G. angustum* Agardh							+			
8, 9	*G. auritum* A.Braun ex Kützing		+				+	+	+		+
10-13	*G. bavaricum* Reichardt & Lange-Bertalot						+	+	+		+
14, 15	*G. brebissonii* Kützing						+	+		+	+
16-18	*G. calcareum* P.T. Cleve					+	+	+			
19-22	*G. calcifugum* Lange-Bertalot & Reichardt							+			
23-24	*G. capitatum* Ehreberg			+			+	+		+	+
25-26	*G. clavatulum* Reichardt	+							+		
27	*G.* cf. *clavatum* Ehrenberg							+			
28-29	*G. designatum* Reichardt						+				
30, 107, 108	*G. dichotomum* Kützing					+					
31, 32	*G. elegantissimum* Reichardt & Lange-Bertalot							+			
33-35	*G. exilissimum* (Grunow) Lange-Bertalot & Reichardt	+	+	+	+		+	+			+
36, 37	*G. geissleriae* Reichardt & Lange-Bertalot							+			
38-42	*G. graciledictum* Reichardt					+					
43-45	*G. hebridense* Gregory	+	+		+		+	+	+		+
46	*G. insigniforme* Reichardt & Lange-Bertalot								+		
47-49	*G. italicum* Ehrenberg							+			
50-52	*G. jadwigiae* Lange-Bertalot & Reichardt		+								
53-56	*G. lateripunctatum* Reichardt & Lange-Bertalot			+	+		+	+	+	+	+
61, 62	*G. linearioides* Levkov (= *Gomphonella*)						+	+	+		+
57-60	*G. minusculum* Krasske				+			+	+		+
63, 64	*G. minutum* (Agardh) Agardh						+	+			+
65-69	*G. olivaceoides* (Hustedt) Lange-Bertalot							+			+
70-74	*G. olivaceum* (Hornemann) Brébisson							+		+	

78-80	*G. pala* Reichardt						+	+	+	+	+
75-77	*G. parvulum* (Kützing) Kützing						+	+		+	+
81-83	*G. pratense* Lange-Bertalot & Reichardt	+						+			+
84-87	*G. procerum* Reichardt & Lange-Bertalot			+			+	+	+		+
88	*G. pseudopusillum* Reichardt					+		+			
89	*G. pseudotenellum* Lange-Bertalot						+	+	+	+	+
90-94	*G. pumilum* (Grunow) Reichardt & Lange-Bertalot	+		+		+	+	+	+		+
95, 96	*G. qii* Juttner										+
97-99	*G. subangustatum* Lange-Bertalot, Cavacini, Tagliaventi & Alfinito										+
100	*G.* cf. *subclavatum* (Grunow) Grunow							+			
101, 102	*G. supertergestinum* Reichardt							+			
104-106	*G. vibrio* Ehrenberg							+	+		
103	*G. vibrio* var. *subcapitatum* (Mayer) Lee							+		+	

Table 3. Occurrence of *Gomphonema* taxa recorded at 10 lakes. 1 - Suchar Wielki (SW), 2 - Wygorzele (WYG), 3 - Wądołek (WAD), 4 - Suchar III (SIII), 5 - Okrągłe (OK), 6 - Białe Wigierskie (BW), 7 - Wigry (W), 8 - Krusznik (K), 9 - Muliczne (M), 10 - Białe Pierciańskie (BP) lakes.


2, 3
4, 5
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16-18
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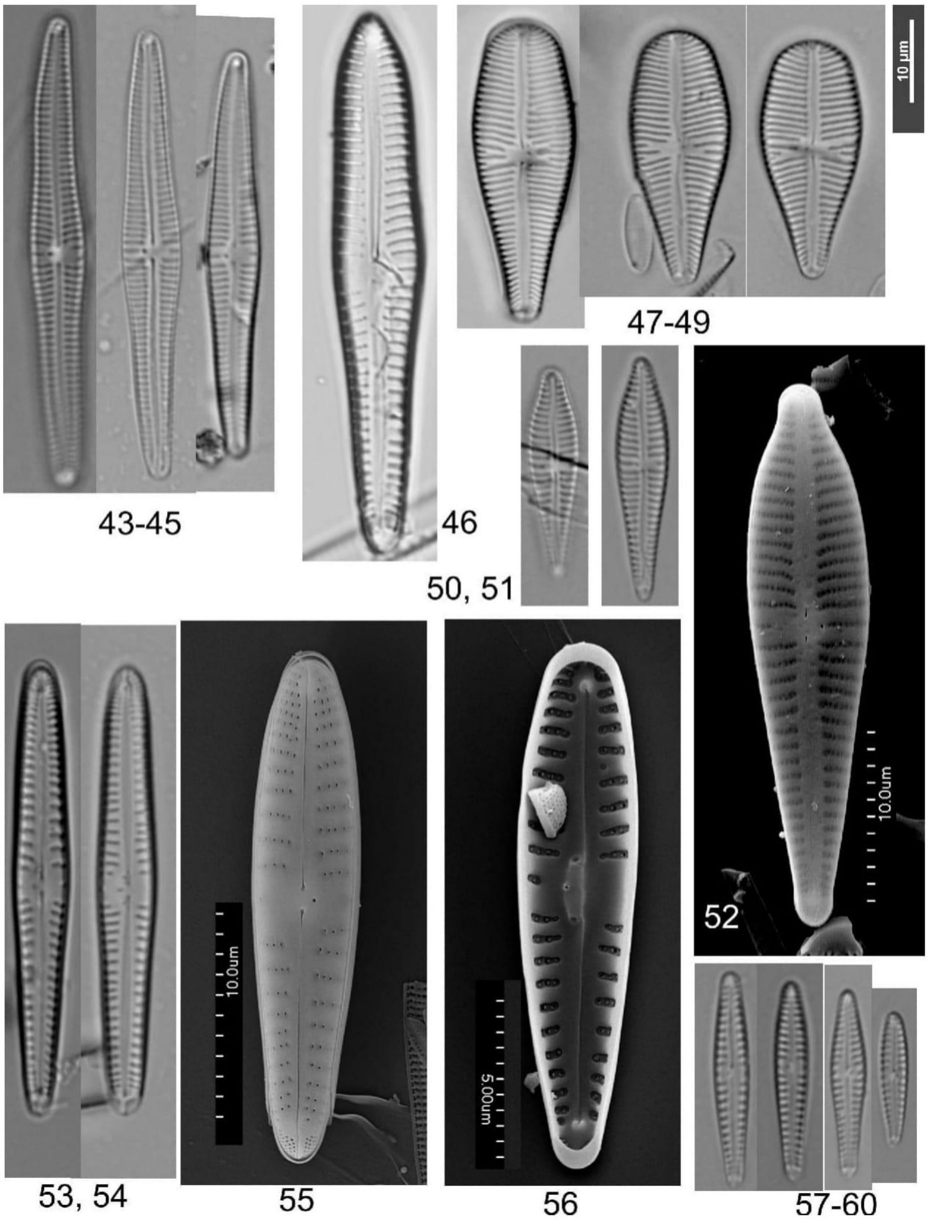
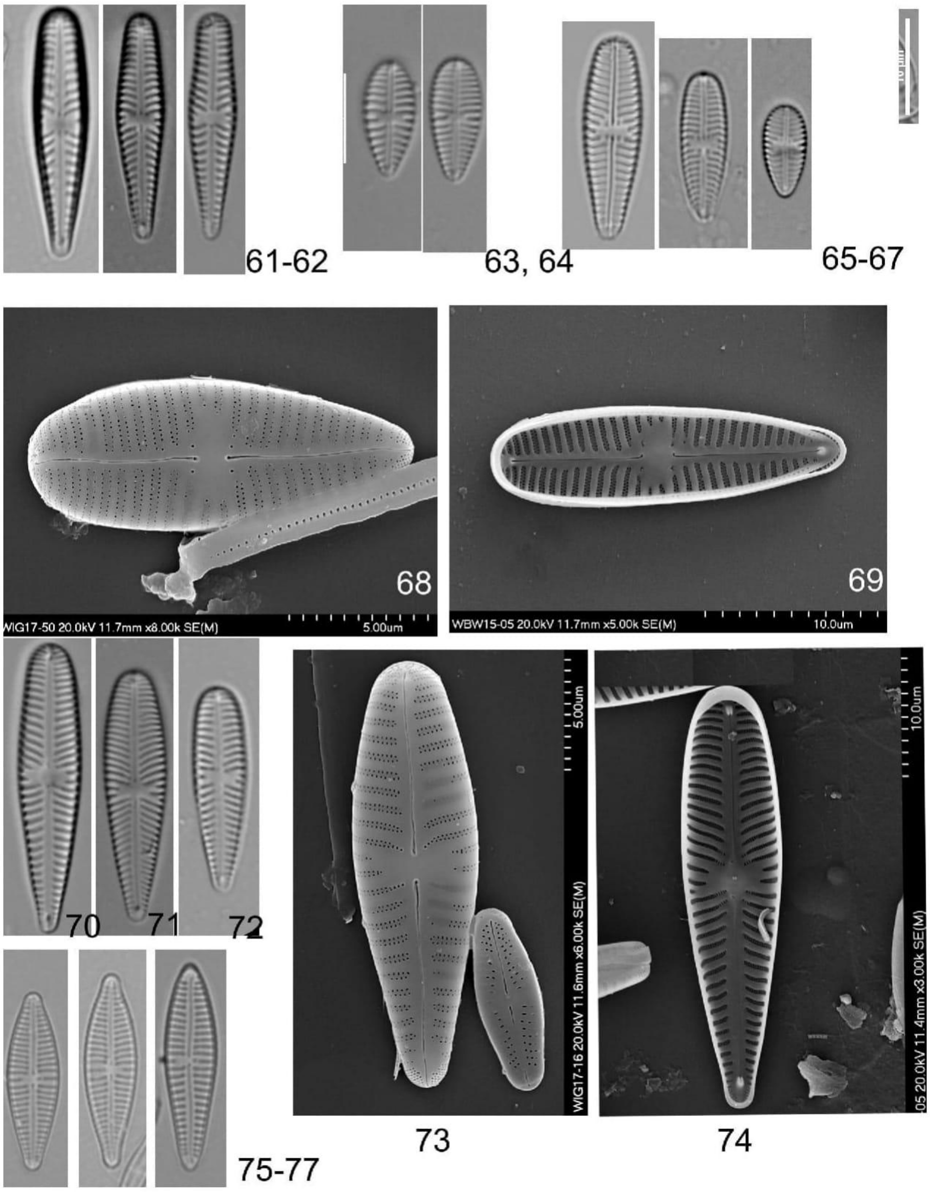
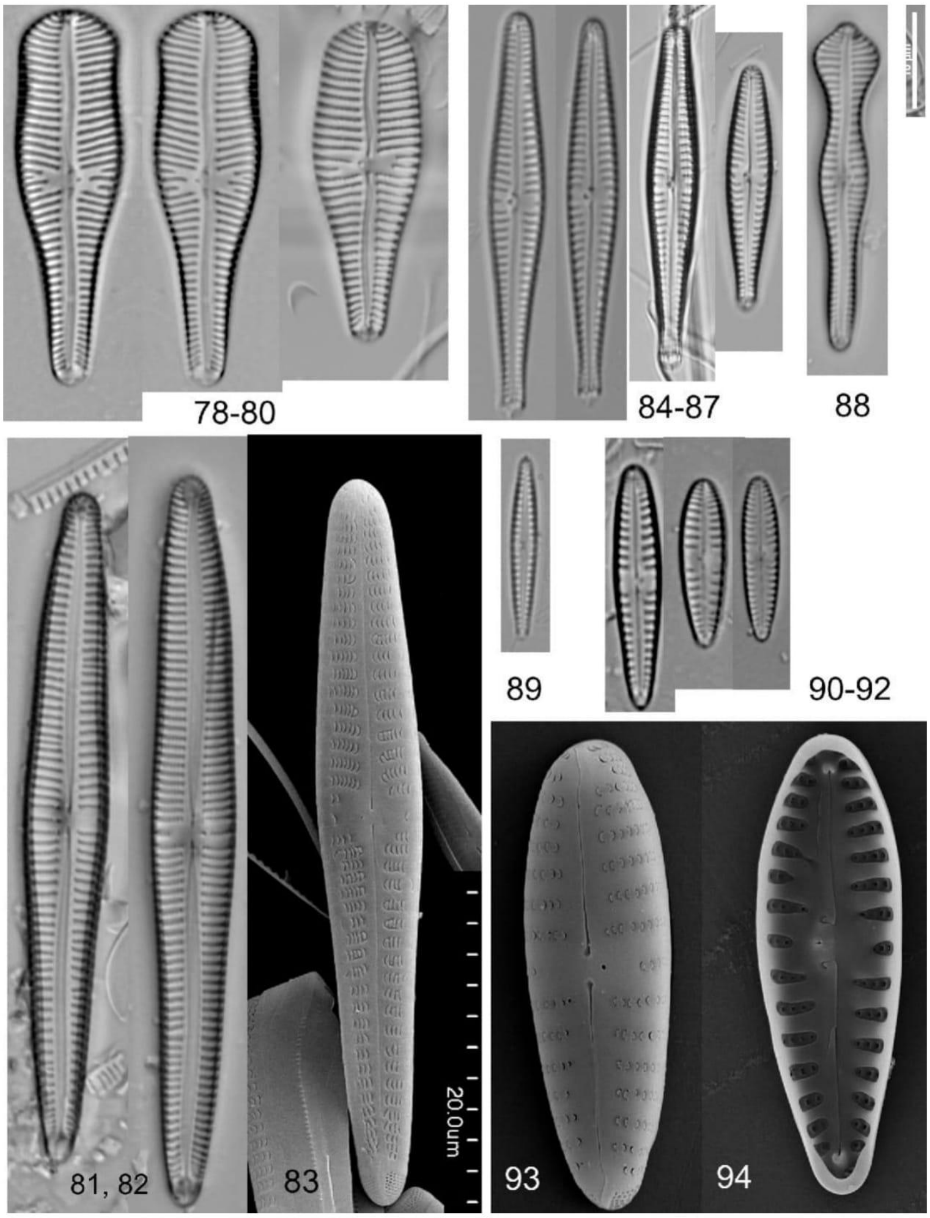
31, 32
36, 37
38-42


10 µm
43-45
46
47-49
50, 51
52
10.0um
53, 54
55
10.0um
56
5.00um
57-60


61-62
63, 64
65-67
68
WIG17-50 20.0kV 11.7mm x8.00k SE(M)
5.00um
69
WBW15-05 20.0kV 11.7mm x5.00k SE(M)
10.0um
70
71
72
5.00um
WIG17-16 20.0kV 11.6mm x6.00k SE(M)
10.0um
05 20.0kV 11.4mm x3.00k SE(M)
73
74
75-77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81, 82
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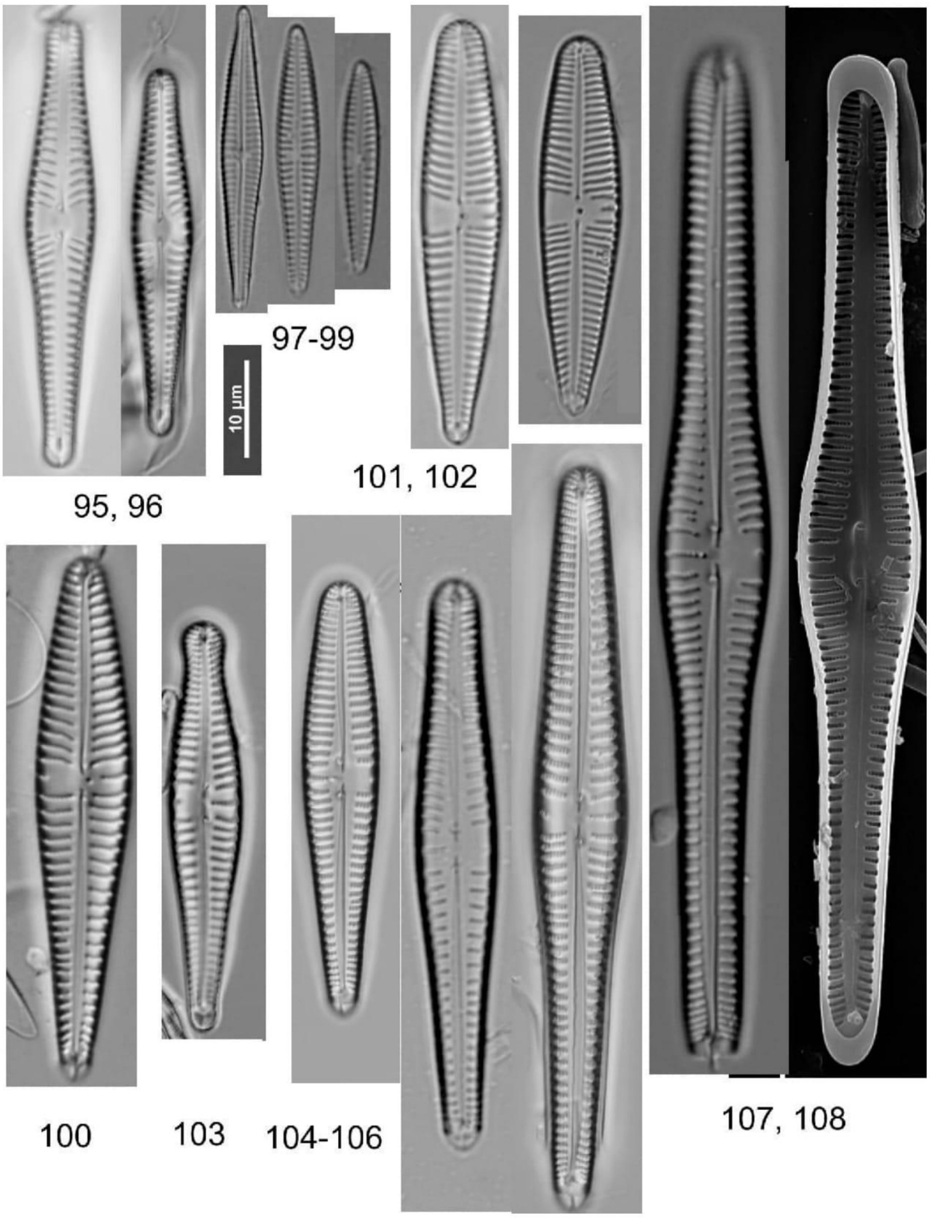
94


97-99
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95, 96
101, 102
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103
104-106
107, 108

SUPLEMENT A:

Różnorodność okrzemek o liczebności powyżej 2% w dziesięciu jeziorach Wigierskiego Parku Narodowego

BP – jez. Białe Pierciańskie, BW – jez. Białe Wigierskie, K – jez. Krusznik, M – jez. Muliczne, SIII – jez. Suchar III, SW – jez. Suchar Wielki, WYG – jez. Wygorzele, WAD – jez. Wądołek, W – jez. Wigry. Środowisko życia (Hab): P–B – planktobentos, B – bentos, Preferencje temperaturowe (T): temp – temperatura umiarkowana, eterm – eurytermia. Preferencje natlenienia i przepływu wody (Oxy): st – woda stojąca, str – woda płynąca, st–str – woda o niskim przepływie. Kwasowość (pH): alb – alkalibionty, alf – alkalifile, ind – szerokie spektrum występowania; acf – acidofile, neu, neutrofile. Stopień zasolenia (Sal): i – oligohaloby – obojętne, hl – halofile, hb – halofoby, mh – mezohaloby. Wskaźniki zanieczyszczenia organicznego według Watanabe (D): sx – saprokseny, es – eurysaprobowe, sp – saprofile. Metabolizm azotu (Aut–Het): ats – taksony azotowo-autotroficzne, tolerujące bardzo małe stężenia azotu związanego organicznie; ate – taksony azotowo-autotroficzne, tolerujące podwyższone stężenia azotu związanego organicznie; hne – fakultatywnie azotowo-heterotroficzne taksony wymagające okresowo podwyższonych stężeń organicznie związanego azotu, hce – fakultatywnie azotowo-heterotroficzne taksony wymagające podwyższonych stężeń organicznie związanego azotu.

Gatunki	O	BW	K	W	BP	M	SIII	SW	WYG	WAD	Hab	T	Oxy	pH	Sal	D	Aut-Het
Achnanthidium affine (Grunow) Czarnecki	+	+	+	+							B	-	str	alf	i	es	-
Achnanthidium minutissimum (Kützing) Czarnecki	+	+	+	+	+	+					P-B	eterm	st-str	ind	i	es	ate
Achnanthidium minutissimum var. *jackii* (Rabenhorst) Lange-Bertalot	+	+	+	+							-	-	-	-	-	-	-
Brachysira neoexilis Lange-Bertalot	+	+	+			+					B	-	-	acf	-	-	-
Brachysira procera Lange-Bertalot & Gerd Moser	+		+								B	-	-	acf	-	-	-
Cocconeis placentula Ehrenberg	+	+	+	+							P-B	temp	st-str	alf	i	es	ate

Cymbella affiniformis Krammer	+	+	+	+							B	-	-	-	-	-	-
Cymbella cymbiformis C.Agardh	+	+	+	+							B	temp	str	ind	i	sx	ats
Cymbella excisa Kützing	+	+	+	+							B	-	-	-	-	-	-
Cymbella lange-bertalotii Krammer	+	+	+	+							-	-	-	-	-	-	-
Cymbella neoleptoceros Krammer	+		+	+							B	temp	st	alf	i	sx	ats
Cymbella proxima Reimer	+	+	+	+							B	-	-	alf	hb	es	-
Cymbella vulgata Krammer	+	+	+	+							B	-	-	ind	-	-	-
Delicata delicatula (Kützing) Krammer	+		+								B, aer	-	str	alf	i	sx	ats
Diatoma ehrenbergii Kützing		+		+							B	-	st-str	alf	i	-	ate
Diatoma tenuis C.Agardh		+		+							P-B	-	st-str	ind	hl	sx	ate
Encyonema cespitosum Kützing	+	+	+	+							B	-	-	-	i	sx	-
Encyonema ventricosum (C.Agardh) Grunow		+		+							B	-	st-str	ind	i	sx	ate
Encyonopsis cesatii (Rabenhorst) Krammer	+	+	+	+							B	-	str	ind	i	es	ats
Encyonopsis krammeri E.Reichardt	+	+	+	+							-	-	-	-	-	-	-
Encyonopsis microcephala (Grunow) Krammer	+	+	+	+	+	+					B	-	str	alf	i	es	ats
Epithemia adnata (Kützing) Brébisson	+	+	+	+							B	temp	st	alb	i	sx	ats

Epithemia sorex Kützing		+		+							B	temp	st-str	alf	i	sx	ats
Eunotia arcubus Nörpel & Lange-Bertalot	+	+	+	+	+						B	-	-	acf	i	sx	-
*Eunotia genuflexa*Nörpel-Schempp 1996							+				B	-	-	acf	-	-	-
Eunotia mucophila (Lange-Bert., Nörpel-Schempp and Alles) Lange-Bert. in Metzeltin et al. 25								+			B	-	-	acf	-	-	-
Eunotia rhomboideaHustedt 195							+		+	+	B	-	-	acf	-	-	-
Fragilaria perminuta (Grunow) Lange-Bertalot	+	+	+	+							-	-	-	alf	-	-	-
Fragilaria subconstricta Østrup		+		+							-	-	-	-	-	-	-
Fragilaria tenera (W.Smith) Lange-Bertalot	+	+	+	+							P-B	-	str	acf	hb	sx	ats
Fragilaria vaucheriae (Kützing) J.B.Petersen	+	+		+							P-B, Ep	-	st-str	alf	i	sx	ate
Fragilariforma mesolepta (Hustedt) Kharitonov		+		+							P-B	-	st-str	alf	i	sx	-
Fragilariforma nitzschioides (Grunow) Lange-Bert. 211								+			-	-	-	-	-	-	-
Gomphonella linearoides (Levkov) R.Jahn & N.Abarca				+							-	-	-	-	-	-	-
Gomphonema auritum A.Braun ex Kützing	+	+	+	+							B	-	-	ind	i	sp	-
Gomphonema bavaricum E.Reichardt & Lange-Bertalot		+	+	+							B	-	-	-	-	-	-

Gomphonema brebissonii Kützing	+		+	+							B	-	st	ind	i	es	-
Gomphonema capitatum Ehrenberg		+	+	+							B	temp	-	alf	i	sx	-
Gomphonema exilissimum (Grunow) Lange-Bertalot & E.Reichardt	+	+	+	+							B	-	str	ind	i	es	Ats
Gomphonema hebridense W.Gregory	+	+	+	+							B	-	-	acf	-	-	-
Gomphonema lateripunctatum E.Reichardt & Lange-Bertalot	+	+	+	+							B	-	str	alf	i	-	Ats
Gomphonema minusculum Krasske	+	+	+	+							B	-	-	-	-	-	-
Gomphonema minutum (C.Agardh) C.Agardh	+	+		+							B	-	-	alf	i	es	-
Gomphonema olivaceoides Hustedt		+		+							B	-	str	ind	i	-	Ats
Gomphonema pala E.Reichardt	+	+	+	+							B	-	-	-	-	-	-
Gomphonema parvulum (Kützing) Kützing	+		+	+							B	temp	str	ind	i	es	Hne
Gomphonema pratense Lange-Bertalot & E.Reichardt				+							-	-	-	-	-	-	-
Gomphonema procerum E.Reichardt & Lange-Bertalot	+	+	+	+							B	-	-	alf	oh	-	-
Gomphonema pseudotenellum Lange-Bertalot	+	+	+	+							B	-	-	-	i	es	-
Gomphonema pumilum (Grunow) E.Reichardt & Lange-Bertalot	+	+	+	+							B	-	-	alf	i	-	-

Gomphonema vibrio Ehrenberg	+	+	+	+							B	-	str	alf	i	es	Ats
Kobayasiella subtilissima (Cleve) Lange-Bert. 1999							+	+	+	+	B	-	-	acf	-	-	-
Mastogloia smithii Thwaites ex W.Smith	+	+	+	+							B	-	-	alf	mh	sx	-
Navicula cryptotenella Lange-Bertalot	+	+	+	+							P-B	-	-	ind	i	es	-
Navicula cryptotenelloides Lange-Bertalot	+	+	+	+							B	-	-	alf	oh	-	-
Nitzschia gracilis Hantzsch 186										+	-	-	-	-	-	-	-
Navicula radiosa Kützing	+	+	+	+							B	temp	st-str	ind	i	es	ate
Navicula subalpina E.Reichardt	+	+	+	+							B	-	-	-	-	es	-
Nitzschia amphibia Grunow	+	+	+	+						+	P-B, S	temp	st-str	alf	i	sp	hne
Nitzschia denticula Grunow	+	+	+	+							-	-	-	-	-	-	-
Nitzschia lacuum Lange-Bertalot		+	+								-	-	str	alf	i	es	ats
Nitzschia palea (Kützing) W.Smith	+	+	+	+							P-B	temp	-	ind	i	sp	hce
Nitzschia sublinearis Hustedt	+	+		+							P-B	-	-	alf	i	es	-
Pseudostaurosira brevistriata (Grunow) D.M.Williams & Round	+	+	+	+							P-B	-	st-str	alf	i	-	-
Rhopalodia gibba (Ehrenberg) O.Müller		+	+	+							B	temp	-	alf	i	es	-

Staurosira construens Ehrenberg	+	+	+								P-B	temp	st-str	alf	i	sx	ats
Staurosirella pinnata (Ehrenb.) D.M.Williams and Round 1987		+									P-B	temp	st-str	alf	i	sx	ats
Tabellaria flocculosa (Roth) Kütz. 1844							+	+	+	+	P-B	eterm	-	ind	-	-	-
Ulnaria acus (Kützing) Aboal	+	+	+	+							P-B	-	st-str	alf	i	es	-
Ulnaria biceps (Kützing) Compère	+	+	+	+							P-B	temp	-	alf	i	-	-
Ulnaria delicatissima (W.Smith) Aboal & P.C.Silva	+	+	+	+							B	-	-	neu	-	es	-

SUPLEMENT B:

Właściwości fizyczne i chemiczne wód w 10 badanych jeziorach Wigierskiego Parku Narodowego BP – jez. Białe Pierciańskie, BW – jez. Białe Wigierskie, O- jez. Okrągłe, K – jez. Krusznik, M – jez. Muliczne, SIII – jez. Suchar III, SW – jez. Suchar Wielki, WYG – jez. Wygorzele, WAD – jez. Wądołek, W – jez. Wigry.

Jeziora	pH	Przewodność	Cl^-	CO_3^{2-}	SO_4^{2-}	NO_3^-	NH_4^+	Mg^{2+}	PO_4^{3-}	Ca^{2+}
		µS/cm	**[mg/L]**							
O	7.68–8.28	338–385	8.45–9.99	103.52–248	12.21–30.92	0.48–1.75	0.04–0.83	11.58–15.58	0.002–0.02	46.81–73.62
M	7.52–8.26	294–342	3.52–3.85	126.70–239	21.35–24.06	0–0.37	0.02–0.22	10.14–14.45	0.002–0.011	40.21–68.25
BP	7.63–8.23	385–401	2.28–2.94	186.77–292	3.15–5.63	0–0.35	0.02–0.32	13.88–18.95	0.002–0.01	48.46–69.45
BW	7.32–8.15	164–185	2.68–3.35	74.81–137.4	6.35–6.81	0.00–0.36	0.01–0.13	4.93–6.53	0.002–0.03	23.85–36.66
K	7.51–8.26	234–342	4.24–4.52	101.49–239	12.36–22.23	0–0.35	0.036–0.23	7.66–14.45	0.002–0.01	30.45–68.25
W	7.56–8.23	350–402	14.47–17.05	135.36–243	21.68–23.57	0–1.26	0.012–0.12	11.80–16.08	0.002–0.03	16.05–45.67
WYG	3.7–6.11	20–25	1.40–2.03	8.44–35.71	0.02–1.23	0.00–0.40	0.20–1.26	0.00–0.49	0.01–0.03	0.85–2.01
WAD	4.65–5.31	20–23	1.27–1.35	17.56–18.60	0.02–0.56	0–0	0.00–0.20	0.77	0.005–0.04	2.67–3.35
SIII	3.6–6.8	22–23	0.86–8.44	9.04–18.45	0.02–1.11	0–0	0.08–0.20	0.00–0.46	0.02–0.05	0.59–1.74
SW	4.60–6.34	15–18	0.97–1.28	8.66–39.66	2.23–2.68	0.00–0.36	0.00–0.00	0.50–0.74	0.01–0.02	1.73–2.75