Community structure and environmental factors affecting diatom abundance and diversity in a Mediterranean climate river system

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HIGHLIGHTS

- Sediment and water chemistry variables increased downstream in both events.
- 110 diatom species across the two study events identified.
- Significant differences observed in diatom community across river zones and events.
- Boron and copper most important variables in structuring diatoms.

GRAPHICAL ABSTRACT
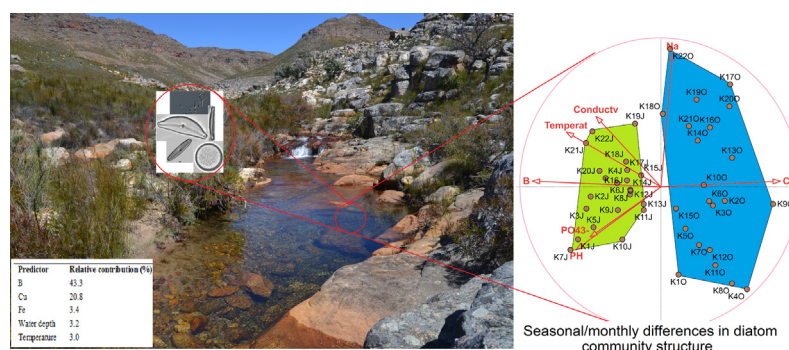
Seasonal/monthly differences in diatom community structure

ARTICLE INFO

ABSTRACT

Mediterranean climate river systems are among the most threatened ecosystems worldwide, due to a long history of anthropogenic impacts and alien invasive species introductions. Many of such rivers naturally exhibit a non-perennial flow regime, with distinct seasonal, inter-annual and spatial heterogeneity. The present study seeks to detect diatom community patterns and to understand the processes that cause these structures in an Austral Mediterranean river system among different months and river sections. In general, most environmental variables showed an increasing trend downstream for both months, with the exception of pH, dissolved oxygen, PO_4^{3-} and substrate embeddedness, which decreased downstream. A total of 110 diatom species between the two study months (October – 106 taxa; January – 78 taxa) were identified, dominated by 30 species with at least >2% abundance. Diatom community structure differed significantly across river zones, while no significant differences were observed between the study months. A boosted regression trees model showed that B (43.3%), Cu (20.8%), Fe (3.4%) and water depth (3.2%) were the most significant variables structuring diatoms. Diatom species communities reflected environmental variables (i.e., sediment and water chemistry) in this Mediterranean climate river system, as sediment metals such as B, Cu and Fe were found to be important in structuring diatom communities. Biotic influences from fish communities had little effect on diversity, but shifted diatom community structure. Therefore, the current study highlights how river systems have complex interactions that play an important role in determining diatom species composition.

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1. Introduction

Rivers are important ecosystems and are home to diverse and abundant animal and plant species, providing vital ecosystem services (Soria et al., 2017; Messager et al., 2021). Lotic freshwater systems are either perennial or temporary, depending on groundwater, surface flow and prevailing environmental conditions. Freshwater ecological communities within these systems are thus adapted to the natural variability through short life spans, mechanisms to resist or avoid desiccation, and high colonisation rates (Feio et al., 2014). Rivers are also critical for humans, being a source of freshwater, food and recreational activities, among other ecosystem services (Dunham et al., 2018). Thus, the conflict between human needs and river system function — given their continuing exposure to human impacts (e.g., considerable changes in sediment delivery, declining water quality, biodiversity loss, flow patterns, morphological modifications) — necessitates management adaptations (Dudgeon et al., 2006; Tan et al., 2017).

Mediterranean climate river systems are among the most threatened ecosystems worldwide, due to a long history of anthropogenic impacts and alien invasive species introductions (Colin et al., 2016). Water diversion, flow regulation, increased salinity, pollution and introduced species have impacted Mediterranean climate river ecosystems over time (Feio et al., 2014). Many of such rivers naturally exhibit a non-perennial flow regime, with distinct seasonal, inter-annual and spatial heterogeneity (Skoulikidis et al., 2017). That is because these systems have climatic conditions that are characterised by marked seasonal differences, such as hot-dry summers and cool-wet winters, sometimes compounded by large inter-annual rainfall variability which affects aquatic biota (Boix et al., 2010; Feio et al., 2014). These Mediterranean river communities therefore show strong inter-annual fluctuations in species richness and composition (Feio et al., 2014). Such systems often additionally have a very complex history and environmental features that have contributed to the high levels of biodiversity and endemism currently hosted (Colin et al., 2016). Thus, organisms in these river systems are often patchily distributed in time and space, rather than homogeneously or randomly, generating considerable spatiotemporal heterogeneity (Bonada et al., 2005; Stomp et al., 2011).

Diatoms are single-celled microscopic organisms belonging to a group of algae, exhibiting a large diversity in terms of taxonomy, morphology and ecology, and are present in most aquatic and humid environments (Round et al., 1990; Liang et al., 2020). Diatoms are cosmopolitan in nature, and are known to be efficient indicators of river ecological quality (Vasselon et al., 2017; Wood et al., 2019; Mangadze et al., 2019a). Bioindicators track environmental variations at differing temporal scales, ranging from weeks (i.e., diatoms, invertebrates) to years (i.e., fish) (Colin et al., 2016; Mangadze et al., 2019b). Diatom community variability is influenced both by natural and anthropogenic factors and species evolutionary adaptations (Stenger-Kovács et al., 2013). Diatoms are accordingly commonly used as biological indicators to assess habitat degradation in river systems, due to their sensitivity to a wide range of environmental stresses (Liu et al., 2020; Gelis et al., 2020). Their use in habitat assessments is supported by their rapid response to changes in environmental conditions, their substantial abundances, and their short generational times (Wood et al., 2019). Their indicator efficiency also relies on their marked taxonomical diversity and their species-specific ecological preferences to different levels of habitat degradation (Pandey et al., 2018; Tan et al., 2017, 2021). However, knowledge gaps in their responses to key environmental variables remain in the Global South and under particular climatic regimes, such as in changeable Mediterranean river systems, hampering understandings of their broad scale efficacy in habitat assessments. As such, it important to analyse diatom species composition in response to environmental factors associated with anthropogenic impacts for better management and protection of aquatic environments. For example, light, nutrients and environmental properties, such as pH and ionic composition, are essential abiotic factors in river ecosystems and their extent and/or availability change spatiotemporally, potentially affecting diatom communities (Stenger-Kovács et al., 2013; Mangadze et al., 2017).

Biotic factors may also be pertinent in influencing diatom communities. In river ecosystems, top predators such as fish often influence primary producer (i.e., diatom (phytoplankton)) abundance and diversity through trophic interactions (Vanni and Layne, 1997; Sommer et al., 2012). Fish-driven trophic cascades can influence primary producers, but with varying strengths (Stevenson et al., 2016; do Nascimento Filho and do Nascimento Moura, 2021) and may arise due to biological invasion (Ellender and Weyl, 2014). For example, diatom community structure is often negatively affected by fish directly or indirectly via reduced abundance and diversity when planktivorous and/or herbivorous fish are abundant. However, diatoms could be positively affected if predatory fish feed on lower-order planktivorous and/or herbivorous fish, reducing their abundance and thus feeding rates on diatoms in consequence (Wasserman et al., 2013). Fish species confer effects through many food web pathways; however, quantifying the importance of these direct and indirect effects is important to understand diatom community structuring and inferences of ecosystem health (Polis et al., 1996; Vanni and Layne, 1997). Therefore, in addition to abiotic factors, diatom species composition is also affected by upper trophic levels, with studies elucidating diatom community structuring by fish fundamental to our understanding of river ecosystem functioning.

The present study seeks to detect diatom community patterns and to understand the processes that cause these structures in an austral Mediterranean river system among different months and river sections, characterised by variable physico-chemical parameters and dominant fish species. Besides the high conservation value of these systems, biodiversity is related to the ecosystem services that these systems provide to society, as well as their ability to cope with multiple stressors. Thus, we expect the abundance and presence of diatoms to be a result of several multi-scale filters, such as the ecological constraints ranging from the landscape to micro-habitat scale (Bonada et al., 2005).

2. Materials and methods

2.1. Study area

The Krom River system is a tributary of the Olifants-Doring River system in the Western Cape Province of South Africa. The river starts at an elevation of ~1500 m in the Cederberg Mountains and flows in an easterly direction for approximately 20 km before joining the Matjies River, a tributary of the Doring River (Fig. 1). The upper river section is narrow (width < 5 m) and shallow (depth < 1 m), comprising of several pools and bedrock steps interspersed by occasional cobble-bed riffles (Marr et al., 2019). The lower river section has deep pools (>3 m), and is isolated during low flow periods (i.e., mid to late summer). The area falls within the Mediterranean climate region, characterised by cold, wet winters and warm, dry summers (Cowling and Holmes, 1992). The mean annual rainfall is estimated at 1000 mm, 500 mm and 250 mm, respectively for the upper, middle and lower reaches. The geology of the river channel is predominantly quartzite sandstones, with minor shale bands of the Nardouw Subgroup, Table Mountain Group and Cape Supergroup (Thamm and Johnson, 2006). The vegetation is dominated by the Cederberg Sandstone Fynbos above the Krom River Cederberg Park, before changing to the northern inland shale band vegetation, just east of a farm complex, and lastly the arid extreme of Cederberg Sandstone Fynbos (Mucina and Rutherford, 2006).

Sampling was conducted in October 2017 and January 2018 (i.e., two seasons). Twenty-two sites were selected on the Krom River system along a zonation of dominant fish presence into three sections i.e., invasive rainbow trout *Oncorhynchus mykiss* (sites K1–K10), native Clanwilliam rock catfish *Austroglanis gilli* (sites K11–K16) and native sawfin *Cheilobarbus serra* (sites K17–K22) zones. It is important to note that in the different fish zones highlighted above, these fish species were the most dominant between both study months in previous studies (Marr et al., 2019), however, other fish species are also present in each zone, such as the invasive bluegill sunfish *Lepomis macrochirus*. Sites K16 – K18 are located within the Cederberg Park camp area and site K22 after the confluence with the
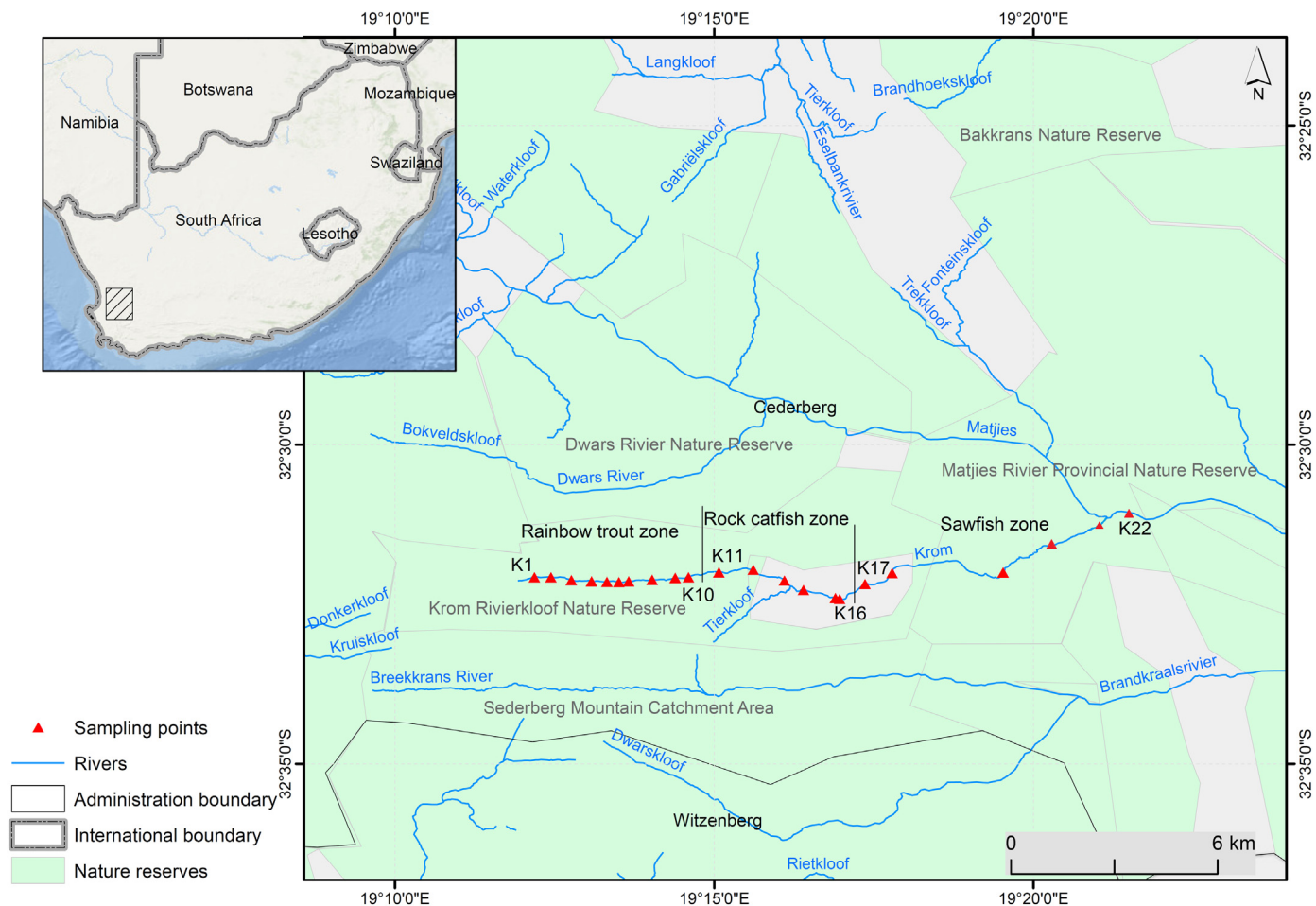
Fig. 1. Location of the study sites in the Krom River, Cederberg Mountain region, South Africa.

Matjies River (Fig. 1; Marr et al., 2019). Approximately 11 km above the Krom River, Cederberg Park is near-pristine and unaffected by human impacts. The river then flows through highly transformed Krom River Cederberg Park, where major abstraction from the Krom River and water diversion to several small reservoirs for irrigation and recreational purposes occurs. Below the park area, the river becomes more seasonal and flows through the near-pristine Matjies River Nature Reserve before joining the Matjies River system.

2.2. Environmental variables sampling and analysis

The key water parameters of conductivity, salinity, pH, temperature and total dissolved solids (TDS) were measured per site and each season using a portable probe PCTestr 35 (Eutech/Oakton Instruments, Singapore). The dissolved oxygen (DO) and turbidity were measured using Per Scientific DO (DO 850045, Taiwan) and turbidity (AL250T-IR, Aqualytic, Germany) meters, respectively. Two 500 mL water samples were collected at each site and season for analysis of nutrients (i.e., ammonium – NH_4^+, phosphates – PO_4^{3-}) via in-field laboratory analyses within 8 h of collection using a HI 83203 multiparameter photometer (Hanna Instruments Inc., Rhode Island). The key habitat characteristics of substrate embeddedness, riparian cover, channel width, water depth and detrital cover, were determined according to Dalu et al. (2017).

Integrated 1.5 kg sediment samples ($n = 2$) were collected from each of the 22 study sites for each sampling event using acid-washed wooden splints and placed in new, labelled ziplock bags for further laboratory analysis. In the laboratory, all sediment samples were oven-dried at 60 °C for 48–72 h before disaggregation and sieving using 0.05 mm mesh size to remove organic matter and large stones. All sediment sample were sent for

analysis at BEMLAB, a South African National Accreditation System (SANAS) certified laboratory. The following were analysed: (i) cation elements: boron (B), calcium (Ca), potassium (K), magnesium (Mg), sodium (Na), (ii) heavy metal: manganese (Mn), copper (Cu), iron (Fe), zinc (Zn) and nutrients: nitrogen (N) and phosphorus (P) (see Dalu et al., 2020a, 2020b for detailed methods).

2.3. Diatom sampling and analysis

Diatom sample collection and analysis were conducted according to Taylor et al. (2005) and Dalu et al. (2020c) by sampling 10 random rocks and/or pebbles per site and season. The collected samples were processed using the hot hydrochloric acid and potassium permanganate method, with diatoms counted (i.e., 300–650 valves per sample) and identified under a phase-contrast Olympus CX light microscope to their lowest taxonomic level using identification guides from Taylor et al. (2007). The following diversity indices were computed: taxa richness, evenness, Shannon-Weiner, and Simpson's indices to assess variations in community structure among sites, zones and seasons.

2.4. Data analyses

Prior to multivariate analyses of environmental data, a test for homogeneity of variance and data normality was conducted and the data were found to violate assumptions of a parametric test. Hence, a non-parametric Kruskal-Wallis test was conducted to assess for differences in environmental variables and diversity indices (i.e., taxa richness, evenness, Shannon-Weiner, Simpson's) data between seasons (i.e., October and January) and river zones (i.e., rainbow trout, rock catfish and sawfish zone) using SPSS

version 16.0 (SPSS Inc., 2007). Distance-based Permutational Analysis of Variance (PERMANOVA; Anderson, 2001) based on Bray-Curtis and Euclidean distance dissimilarities and 9999 permutations with Monte Carlo tests were performed to analyse differences in diatom communities data among months and river sections using PERMANOVA + for PRIMER version 6 (Anderson et al., 2008).

Differences in environmental variables and diatom species composition between months and zones were tested using a two-way analysis of similarity (ANOSIM) with 9999 permutations using PRIMER v6 add-on package PERMANOVA + (Anderson et al., 2008; Clarke, 1993). The ANOSIM statistic R was based on the difference of mean ranks between groups and within groups. It varies between −1 and +1, with value 0 indicating random grouping.

We further employed a Boosted Regression Tree (BRT) model to assess the relationships between environmental variables and diatom species richness. The BRT uses two algorithms, namely boosting and regression (Schapire, 2003; Elith et al., 2008). The fitting of multiple trees in BRT overcomes the biggest drawback of single tree models, and the BRT model was carried out in the R statistical package (R Core Team, 2020), using the BRT extension gbm package (i.e., gbm.step and gbm.simplify function) (Ridgeway, 2006; Elith et al., 2008). A Type I error of 0.05 was used in all analyses.

3. Results

3.1. Environmental variables

In general, most environmental variables showed an increasing trend downstream for both months, with the exception of pH, dissolved oxygen, PO_4^{3-} and substrate embeddedness, which decreased downstream (Table 1). Most of the water and habitat characteristics (i.e., temperature, pH, DO, conductivity, TDS, salinity, turbidity, riparian cover, detrital cover, water depth, substrate embeddedness, NH_4^+) and several sediment variables (i.e., N, Mg, Na, Zn) were found to be significantly different (Kruskal-Wallis (KW), $p < 0.05$) among the river zones, whereas significant seasonal differences (KW, $p < 0.05$) were observed for water and habitat characteristics (i.e., temperature, pH, conductivity, salinity, turbidity, water depth, riparian cover, detrital cover,

Table 2
Kruskal-Wallis results of the measured environmental variables for the different fish zones and months in the Krom River system, South Africa. Significant terms are emboldened.

Variable	Zone		Season	
	H	p	H	p
Water				
Temperature	**29.297**	**<0.001**	**45.273**	**<0.001**
pH	**36.117**	**<0.001**	**14.808**	**<0.001**
Dissolved oxygen	**24.772**	**<0.001**	0.582	0.446
Conductivity	**40.909**	**<0.001**	**5.149**	**0.023**
TDS	**53.158**	**<0.001**	0.585	0.445
Salinity	**44.657**	**<0.001**	**8.549**	**0.003**
Turbidity	**12.313**	**0.002**	**49.161**	**<0.001**
Water depth	2.624	0.269	**8.267**	**0.004**
Riparian cover	**6.815**	**0.033**	**5.423**	**0.020**
Channel width	2.179	0.336	1.672	0.196
Detrital cover	**9.065**	**0.011**	**14.597**	**<0.001**
Substrate embeddedness	**11.443**	**0.003**	0.108	0.743
PO_4^{3-}	0.163	0.922	**11.241**	**0.001**
NH_4^+	**6.184**	**0.045**	1.930	0.165
Sediment				
N	**8.188**	**0.017**	3.664	0.056
K	2.097	0.350	**4.302**	**0.038**
Ca	3.214	0.200	**5.500**	**0.019**
Mg	**6.963**	**0.031**	0.478	0.489
Na	**20.429**	**<0.001**	0.372	0.542
Mn	2.329	0.312	**7.677**	**0.006**
Fe	0.856	0.652	0.005	0.944
Cu	2.275	0.321	**30.715**	**<0.001**
Zn	**6.733**	**0.035**	2.268	0.132
B	0.027	0.986	**34.796**	**<0.001**

PO_4^{3-}) and several sediment variables (i.e., K, Ca, Mn, Cu, B) (Table 2). Sediment P was below detection limit among all sites and seasons.

3.2. Diatom community structure and relationship with environmental variables

A total of 110 diatom species between the two study months (October – 106 taxa; January – 78 taxa) were identified, dominated by 30 species with

Table 1
Mean (± standard deviation) of environmental variables measured from the Krom River system, South Africa. Abbreviations: TDS – total dissolved solids, PO_4^{3-} – phosphates, NH_4^+ − ammonium.

Variable	Units	October			January		
		Rainbow trout	Rock catfish	Sawfish	Rainbow trout	Rock catfish	Sawfish
Water and habitat characteristics							
Temperature	°C	15.1 ± 0.4	16.3 ± 0.8	18.0 ± 0.2	18.5 ± 0.6	19.6 ± 0.3	23.2 ± 0.4
pH		6.8 ± 0.2	6.7 ± 0.6	6.3 ± 0.3	8.1 ± 0.5	6.9 ± 0.3	6.5 ± 0.1
Dissolved oxygen	mg L^{-1}	7.7 ± 0.2	7.5 ± 0.3	7.0 ± 0.2	7.5 ± 0.6	7.0 ± 0.7	5.9 ± 1.2
Conductivity	μS cm^{-1}	27.5 ± 8.1	30.7 ± 8.2	52.6 ± 29.8	31.3 ± 5.6	37.4 ± 6.3	117.1 ± 5.0
TDS	mg L^{-1}	19.5 ± 5.6	23.1 ± 5.3	37.4 ± 21.2	19.7 ± 3.4	26.2 ± 3.9	81.4 ± 3.6
Salinity	ppm	0.02 ± 0.003	0.02 ± 0.002	0.03 ± 0.02	0.01 ± 0.001	0.01 ± 0.001	0.04 ± 0.001
Turbidity	NTU	0.8 ± 0.3	1.3 ± 0.5	2.3 ± 0.6	0.8 ± 1.3	3.7 ± 4.6	4.3 ± 1.7
Water depth	cm	47.8 ± 0.3	61.1 ± 0.5	76.3 ± 0.6	32.0 ± 1.4	29.9 ± 4.6	28.3 ± 1.7
Riparian cover	%	27.4 ± 31.5	37.0 ± 32.4	45.3 ± 31.2	20.6 ± 14.0	15.0 ± 16.2	15.0 ± 9.0
Channel width	cm	312.2 ± 101.3	271.1 ± 132.8	279.2 ± 97.2	301.0 ± 144.9	388.3 ± 89.4	563.6 ± 135.6
Detrital cover	%	2.7 ± 2.0	4.2 ± 4.9	37.5 ± 28.9	30.5 ± 20.9	33.3 ± 19.7	45.0 ± 22.0
Substrate embeddedness		4.2 ± 2.0	3.5 ± 1.4	2.7 ± 1.9	4.1 ± 0.7	3.5 ± 1.8	2.2 ± 1.8
PO_4^{3-}	mg L^{-1}	0.6 ± 0.9	0.3 ± 0.7	0.3 ± 0.7	1.3 ± 1.5	0.9 ± 0.5	1.1 ± 0.4
NH_4^+	mg L^{-1}	0.6 ± 1.8	0.1 ± 0.1	0.4 ± 0.5	0.2 ± 0.3	0.2 ± 0.2	0.5 ± 0.3
Sediment							
N	%	0.03 ± 0.01	0.03 ± 0.004	0.04 ± 0.01	0.02 ± 0.01	0.04 ± 0.03	0.03 ± 0.01
K	%	0.01 ± 0.01	0.02 ± 0.0	0.02 ± 0.01	0.02 ± 0.01	0.10 ± 0.09	0.03 ± 0.03
Ca	%	0.001 ± 0.003	0.002 ± 0.004	0.01 ± 0.02	0	0	0
Mg	%	0.001 ± 0.003	0.003 ± 0.01	0.01 ± 0.01	0.0 ± 0.0	0.003 ± 0.01	0.01 ± 0.01
Na	mg kg^{-1}	412.8 ± 47.5	416.0 ± 49.8	568.4 ± 93.8	375.7 ± 96.4	512.0 ± 207.1	1009 ± 156.0
Mn	mg kg^{-1}	5.7 ± 3.4	4.1 ± 2.6	23.0 ± 40.0	17.1 ± 16.5	17.8 ± 22.5	21.8 ± 21.8
Fe	mg kg^{-1}	1081 ± 583.5	1169 ± 795.8	1585 ± 1141	1101 ± 275.7	1333 ± 734.6	816.0 ± 411.5
Cu	mg kg^{-1}	0.6 ± 0.4	2.1 ± 1.5	6.5 ± 3.3	451.3 ± 981.5	40.5 ± 40.2	88.8 ± 170.3
Zn	mg kg^{-1}	2.8 ± 1.3	3.9 ± 3.0	15.0 ± 11.1	3.2 ± 3.5	2.5 ± 1.9	96.0 ± 144.0
B	mg kg^{-1}	30.5 ± 11.9	29.3 ± 16.9	38.1 ± 25.6	0.2 ± 0.6	0.2 ± 0.4	2.0 ± 4.9

at least >2% abundance (Table 3, Table S1). The most common taxa were *Diatoma vulgaris*, *Eunotia formica*, *Eunotia minor*, *Navicula radiosa*, *Pinnularia viridoformis* and *Tabellaria flocculosa*. Based on PERMANOVA analysis, significant differences were observed in diatom community structure across the river zones (Pseudo-F = 3.527, p = 0.0001) and months (Pseudo-F = 3.527, p = 0.0001), with significant pair-wise differences being observed between the rainbow trout and rock catfish zones (t = 1.594, p = 0.0034). The sawfish zone had the highest taxa richness, with diatom diversity indices being relatively high in the rock catfish zone (Table 3). No significant differences (KW, p > 0.05) were observed among the river zones (all diversity indices) and for Simpson's diversity index between seasons (KW, H = 2.511, p = 0.113), with significant seasonal differences being observed for taxa richness (KW, H = 31.715, p < 0.001), Shannon-Weiner (KW, H = 13.067, p < 0.001) and Evenness (KW, H = 21.819, p < 0.001) diversity indices.

Based on the ANOSIM results, diatom community structure differed significantly across river zones (R = 0.245, p = 0.010), while no significant differences were observed between the study months (R = 0.030, p = 0.283). No significant differences (p > 0.05) were, however, observed in diatom community structure based on pair-wise comparisons among the different river zones. The boosted regression trees (BRT) model predictive performance relating diatom species richness to environment variables was as follows: 3780 trees fitted (boosted models); 53.4 and 0.09 total deviance and mean residual of the model, respectively; 7.18 ± 2.23 estimated cross-validated (CV) residual deviance ± standard error; and CV correlation of 0.94 ± 0.02. According to the BRT model, all variables were retained as significant terms relating diatom species richness to environment variables, with the first eleven variables (i.e., B (43.3%), Cu (20.8%), Fe (3.4%), water depth (3.2%), temperature (3.0%), PO_4^{3-} (2.9%), Mn (2.6%), detrital cover (2.6%), Zn (2.4%), Na (2.3%), NH_4^+ (1.9%)) accounting for ~90% of the species richness variation, with B and Cu concentrations being the most significant (Table 4). Relationships

Table 4

Predictor variable contribution to boosted regression tree models relating diatom species richness to environmental variables learning rate of 0.05 and tree complexity of 5. Environmental variables are sorted in order of decreasing diatom richness contribution.

Predictor	Relative contribution (%)	Predictor	Relative contribution (%)
B	43.3	Riparian cover	1.8
Cu	20.8	TDS	1.8
Fe	3.4	Turbidity	1.5
Water depth	3.2	Channel width	1.4
Temperature	3.0	Salinity	1.0
PO_4^{3-}	2.9	pH	1.0
Mn	2.6	Substrate embeddedness	1.0
Detrital cover	2.6	Conductivity	0.9
Zn	2.4	DO	0.9
Na	2.3	K	0.1
NH_4^+	1.9	N	0.1

fitted by the BRT models for the environmental variables and diatom species richness are highlighted in Fig. 2. Predicted species richness had an approximately sigmoid shaped (i.e. positive) relationship with B concentration, with high values occurring over a wide range characterised by low water depth, Zn, Mn and Fe, and high Cu and detrital cover. A more muted variation occurred in relation to water temperature, Zn, Mn, detrital cover and Fe concentrations. (Fig. 2). A generally high diatom species richness was predicted for seasonal sites with high Cu, detrital cover and phosphates.

4. Discussion

The current study demonstrated that the Krom River diatom community was largely structured by sediment chemistry variables, particularly B, Cu
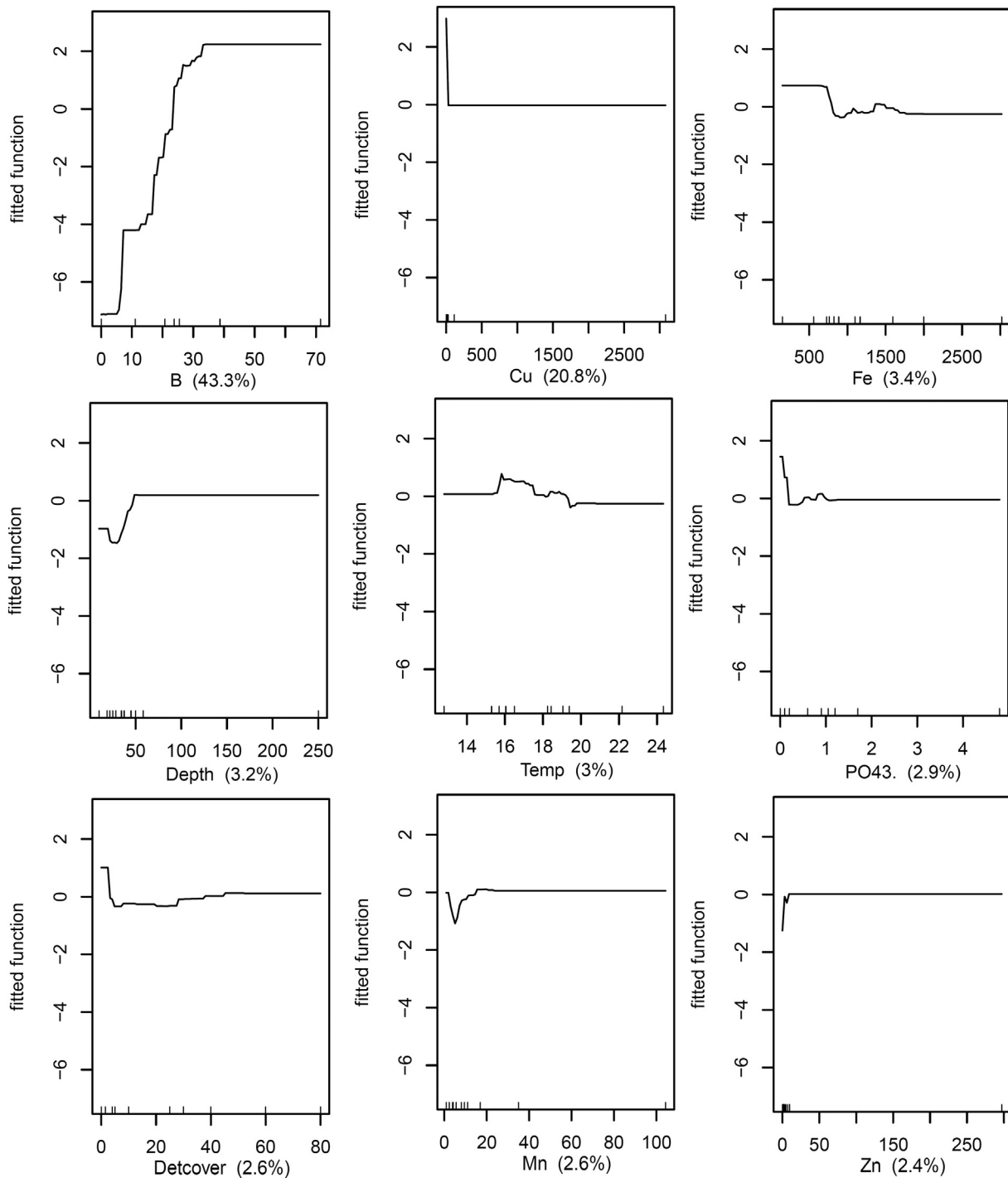
Table 3

Dominant diatom species relative percentage abundances ± standard deviation (species with >2%) collected in three river zones (rainbow trout zone, rock catfish zone, sawfish zone) between the two study months (October 2017, January 2018) along the Krom River system, South Africa.

Species	October			January		
	Rainbow trout	Rock catfish	Sawfish	Rainbow trout	Rock catfish	Sawfish
Cymbella cymbiformis		2.1 ± 4.3	2.0 ± 3.2		1.4 ± 1.4	0.2 ± 0.4
Cymbella tumida	1.5 ± 4.7	1.2 ± 1.5	1.8 ± 3.0	0.1 ± 0.2	0.3 ± 0.6	0.2 ± 0.3
Diatoma vulgaris	1.1 ± 3.5	10.8 ± 8.8	0.1 ± 0.2	1.5 ± 1.9	5.8 ± 6.3	1.3 ± 1.5
Eunotia formica	5.4 ± 10.0	12.8 ± 8.8	4.5 ± 8.8	10.9 ± 8.4	8.9 ± 4.4	8.7 ± 6.0
Eunotia minor	3.7 ± 5.4	15.8 ± 10.9	7.2 ± 7.0	3.2 ± 3.4	12.4 ± 7.2	2.7 ± 1.7
Eunotia rhomboidea	14.5 ± 12.8	6.0 ± 10.1	6.1 ± 7.0	3.3 ± 3.2	7.5 ± 4.7	9.9 ± 7.0
Fragilaria sp.	2.8 ± 6.9		0.5 ± 0.6	1.2 ± 3.2	0.3 ± 0.6	0.9 ± 1.2
Fragilaria ulna	5.0 ± 6.4	0.3 ± 0.8	2.1 ± 2.5	0.7 ± 1.1	0.1 ± 0.2	0.9 ± 1.2
Frustalia rostrata	2.3 ± 4.0	0.2 ± 0.5		0.5 ± 1.1	0.1 ± 0.4	0.1 ± 0.2
Frustalia saxonica	2.6 ± 5.5	3.3 ± 5.4	0.4 ± 0.6	0.5 ± 1.1	0.1 ± 0.2	0.1 ± 0.3
Frustalia magaliesmontana		0.6 ± 1.4	3.1 ± 5.7	0.7 ± 1.5	0.2 ± 0.5	1.1 ± 2.0
Gomphonema acuminatum			0.1 ± 0.2	3.4 ± 5.8	1.3 ± 3.2	0.4 ± 0.9
Gomphonema loticullum				3.0 ± 4.9	1.4 ± 3.0	
Gomphonema parvulum	0.5 ± 1.4		2.2 ± 5.3	4.8 ± 6.5	1.1 ± 2.4	0.1 ± 0.2
Gomphonema venusta	2.2 ± 4.1			4.8 ± 7.5	1.0 ± 2.4	0.3 ± 0.8
Navicula heimansoides	2.4 ± 5.5	2.0 ± 4.9	2.0 ± 4.8	0.3 ± 0.6	0.7 ± 1.4	1.1 ± 1.9
Navicula ranomafenensis	4.5 ± 5.6	1.2 ± 2.0	8.7 ± 6.4		1.2 ± 2.4	0.7 ± 1.7
Navicula radiosa	10.0 ± 7.5	8.5 ± 8.4	6.3 ± 4.6	5.1 ± 5.7	4.8 ± 4.6	1.8 ± 2.1
Navicula sp. 1	2.5 ± 3.4	0.1 ± 0.2	2.5 ± 2.7	0.5 ± 1.0	0.1 ± 0.1	0.9 ± 1.2
Navicula sp. 2	0.4 ± 1.1			2.4 ± 5.5	0.1 ± 0.2	
Navicula subrhynchocephala				2.0 ± 3.6		0.4 ± 0.9
Navicula zanonii	0.2 ± 0.5	0.7 ± 1.8		2.0 ± 3.4	0.2 ± 0.4	1.3 ± 2.6
Nitzschia sigma	2.0 ± 2.2	0.2 ± 0.3	1.6 ± 1.6	0.2 ± 0.3	0.1 ± 0.2	0.3 ± 0.5
Pinnularia acrosphaeria				3.0 ± 5.9	1.3 ± 2.1	0.1 ± 0.2
Pinnularia divergens var. *undulata*	2.1 ± 4.4	0.7 ± 0.8	1.6 ± 3.5	0.6 ± 1.7	0.6 ± 1.3	0.3 ± 0.5
Pinnularia viridoformis	2.8 ± 3.5	3.2 ± 3.8	4.8 ± 5.5	2.1 ± 4.4	2.0 ± 2.8	2.6 ± 2.8
Staurosira elliptica	2.4 ± 6.1	2.9 ± 7.1		0.5 ± 1.2		0.1 ± 0.3
Surirella sp.	0.1 ± 0.3			3.8 ± 11.6		0.1 ± 0.3
Tabellaria flocculosa	5.0 ± 11.5	7.9 ± 12.3	10.3 ± 13.2	4.7 ± 6.1	8.7 ± 10.5	25.5 ± 17.6
Tryblionella apiculata	0.1 ± 0.3			2.1 ± 6.6	4.8 ± 11.9	3.3 ± 8.0
Taxa richness	27.1 ± 5.4	27.2 ± 4.1	28.2 ± 5.7	13.4 ± 2.7	13.8 ± 2.2	16.2 ± 3.3
Shannon-Weiner	2.6 ± 0.3	2.6 ± 0.2	2.4 ± 0.3	2.2 ± 0.3	2.1 ± 0.2	2.3 ± 0.2
Simpson	0.89 ± 0.04	0.90 ± 0.03	0.83 ± 0.07	0.86 ± 0.05	0.84 ± 0.04	0.87 ± 0.03
Evenness	0.50 ± 0.08	0.52 ± 0.10	0.42 ± 0.06	0.71 ± 0.08	0.61 ± 0.10	0.63 ± 0.08

Fig. 2. Variation in diatom species richness predicted by a boosted regression trees (BRT) model for the first 9 environmental variables, and using environmental characteristics as predictors.

and Fe which accounted for 67.5% of the variation. This provides valuable information on factors influencing diatom composition within Mediterranean climate river systems in the Global South. Moreover, the monthly diatom composition differed due to underlying environmental variables, but also potentially as a result of the different fish species present in each zone and their respective feeding preferences. The present study therefore showed the existence of a strong spatial and temporal component in diatom community structuring, but with no distinctly different diatom diversities among river zones, as highlighted by our analyses. The sediment chemistry variables were generally low, as expected due to the near pristine environment where the river is located and the underlying area geology (Thamm

and Johnson, 2006). A number of diatom species were common among zones and seasons, but there were differences in their frequencies, potentially due to fish community composition that could have exerted a top-down effect on the system. Our results are similar to do Nascimento Filho and do Nascimento Moura (2021), in the respect that rainbow trout, rockfish and sawfish had a strong predatory role affecting macroinvertebrates, which might have affected diatom community composition and abundances through cascade effects. A lack of differences among zones considering diversity indices suggests, however, that the effects of these alien and native fish species are consistent with regards to the richness and evenness of diatom communities. Macroinvertebrates have been shown to efficiently

reduce diatom species, especially in the absence of top predators which consume them (Vanni and Layne, 1997; Mermillod-Blondin et al., 2020).

While diatom community structure differed significantly, certain similarities in diatom diversity could be due to differences in structure (i.e., species composition, but similar diversity levels therein). As a consequence, the indicator taxa for the near-natural streams proved to be type-specific. Approximately 110 diatom species were observed, which were dominated by weakly or poor-to-moderate electrolyte species, such as *Eunotia formica*, *Diatoma vulgaris*, *Eunotia rhomboidea* and *Tabellaria flocculosa*. This was further supported by the weak ion and conductivity concentrations recorded within the study sites and several other studies (see Table 1; De Fabricius et al., 2003; Taylor et al., 2007). Diatom species diversity was relatively similar across river zones, but was significantly different between the study months, with lower species richness during the January month in particular. This could be attributed to changes water levels which caused changes in the water and sediment chemistry. Death et al. (2009), Boix et al. (2010) and Dalu et al. (2016) highlighted that hydrological alterations accounted for ecosystem community composition, structure and functioning changes.

Diatom development depends on various environmental variables such as metals. However, when these metal micronutrients are present at an optimal level, diatoms exhibit a strong fitness and grow at their fastest rate (Masmoudi et al., 2013). Boosted regression tree results indicated a highly predictable relationship between diatom species richness and environmental (i.e., B, Cu, Fe, water depth, temperature) variables. Sediment B concentration was the strongest predictor of diatom species richness, with predicted richness peaking at a concentration ~ 30 mg kg^{-1}. A positive association between diatom species richness and other environmental variables (i.e., water depth, Mn and Zn concentration) was also observed, with maximum diatom richness being recorded at sites with low water depth, Zn, Fe and Mn concentrations, and high detrital cover, Cu and PO$_4^{3-}$ concentrations. Lewin (1966) and Loomis and Durst (1992) highlighted that all diatoms require B, as do all higher plants, and it is thus considered as an essential nutrient for diatom growth (i.e., cell wall structure and function) and physiological needs. Thus, B has been shown to play an important role in the general structure and function of diatom plasma membranes, where it influences membrane domain stability through binding with the glycoproteins, glycolipids and phosphoinositides (Brown et al., 2002).

Similarly, diatoms require small amounts of metal ions, such as Ca, Cu, Fe, K, Mg, Mn and Zn, which are necessary for development and growth, and hence are crucial for cell physiology (Masmoudi et al., 2013). Studies have shown that *Cyclotella* spp. and *Fragilaria* spp. disappeared due to Cu pollution, whereas other species presented various deformations in response to Cu and Fe. In contrast, *Achnanthes* spp. were able to accumulate and tolerate Cu over time (Ruggiu et al., 1998; Lavoie et al., 2017; Moreno et al., 2020). Water temperature was found to be a further significant factor affecting diatom community structuring within the Krom River system across seasons and/or river sections. With monthly changes, there were significant zonal and seasonal differences in riparian cover and water temperature, with the riparian cover reducing in January and leading to increased water temperatures. Ingebrigtsen et al. (2016) and Xiao et al. (2018) highlighted that increased light intensity and water temperatures normally result in changes to diatom species composition, which could explain the differences in species richness observed here seasonally.

In conclusion, diatom species communities reflected environmental variables (i.e., sediment and water chemistry) in this Mediterranean climate river system, as sediment metals such as B, Cu and Fe were found to be important in structuring diatom communities. Biotic influences from fish communities had little effect on diversity, but shifted community structure. Therefore, the current study highlighted how Mediterranean river systems in the Global South have complex interactions that played an important role in determining the diatom species composition. More studies are required to fully understand how environmental variables and fish compositions shape diatom communities in these systems, with the aim to improve biomonitoring efforts.

Supplementary data to this article can be found online at https://doi.org/10.1016/j.scitotenv.2021.152366.

CRediT authorship contribution statement

Tatenda Dalu: Conceptualization, Investigation, Methodology, Data curation, Formal analysis, Funding acquisition, Writing – original draft, Writing – review & editing. **Ross N. Cuthbert:** Methodology, Writing – original draft, Writing – review & editing. **Olaf L.F. Weyl:** Supervision, Funding acquisition, Project administration. **Ryan J. Wasserman:** Conceptualization, Funding acquisition, Writing – original draft, Writing – review & editing.

Declaration of competing interest

All authors declare no conflict or financial interests exist for the manuscript.

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