UNIVERSITY OF LEEDS

This is a repository copy of *Declining glacier cover threatens the biodiversity of alpine river diatom assemblages*.

White Rose Research Online URL for this paper:
http://eprints.whiterose.ac.uk/136149/

Version: Accepted Version

Article:

Fell, SC, Carrivick, JL orcid.org/0000-0002-9286-5348, Kelly, MG et al. (2 more authors) (2018) Declining glacier cover threatens the biodiversity of alpine river diatom assemblages. Global Change Biology, 24 (12). pp. 5828-5840. ISSN 1354-1013

https://doi.org/10.1111/gcb.14454

© 2018 John Wiley & Sons Ltd. This is the peer reviewed version of the following article: Fell, S, Carrivick, J, Kelly, M et al. (2 more authors) Declining glacier cover threatens the biodiversity of alpine river diatom assemblages. Global Change Biology. ISSN 1365-2486 (In Press), which has been published in final form at https://doi.org/10.1111/gcb.14454. This article may be used for non-commercial purposes in accordance with Wiley Terms and Conditions for Use of Self-Archived Versions.

Reuse
Items deposited in White Rose Research Online are protected by copyright, with all rights reserved unless indicated otherwise. They may be downloaded and/or printed for private study, or other acts as permitted by national copyright laws. The publisher or other rights holders may allow further reproduction and re-use of the full text version. This is indicated by the licence information on the White Rose Research Online record for the item.

Takedown
If you consider content in White Rose Research Online to be in breach of UK law, please notify us by emailing eprints@whiterose.ac.uk including the URL of the record and the reason for the withdrawal request.

White Rose
university consortium
Universities of Leeds, Sheffield & York

eprints@whiterose.ac.uk
https://eprints.whiterose.ac.uk/

Declining glacier cover threatens the biodiversity of alpine river diatom assemblages

Sarah C. Fell[1], Jonathan L. Carrivick[1], Martyn G. Kelly[2], Leopold Füreder[3], Lee E. Brown[1*]

[1]School of Geography and water@leeds, University of Leeds, Woodhouse Lane, Leeds, LS2 9JT, UK.
[2]Bowburn Consultancy, 11 Monteigne Drive, Durham, DH6 5QB, UK.
[3]Institute of Ecology, University of Innsbruck, Innrain 52, 6020 Innsbruck, Austria.
*Corresponding author
E: l.brown@leeds.ac.uk
T: +44(0) 113 34 33302

Key words:
Algae, Alps, climate change, diatom, glacier retreat, mountain

Paper type:
Primary Research Article

Running head:
Alpine river diatoms

Abstract

Climate change poses a considerable threat to the biodiversity of high altitude ecosystems worldwide, including cold-water river systems that are responding rapidly to a shrinking cryosphere. Most recent research has demonstrated the severe vulnerability of river invertebrates to glacier retreat but effects upon other aquatic groups remain poorly quantified. Using new datasets from the European Alps, we show significant responses to declining glacier cover for diatoms, which play a critical functional role as freshwater primary producers. Specifically, diatom α-diversity and density in rivers presently fed by glaciers will increase with future deglaciation, yet β-diversity within and between-sites will reduce because declining glacier influence will lower the spatiotemporal variability of glacier cover and its associated habitat heterogeneity. Changes in diatom assemblage composition as glacier cover declined were associated strongly with increasing river-bed stability and water temperature. At the species level, diatoms showed a gradation of responses; for example, Eunotia trinacria, found exclusively at river sites with high (≥ 52 %) catchment glacier cover, may be affected negatively by ice loss. Conversely, seven taxa confined to sites with no glacier cover, including Gomphonema calcareum, stand to benefit. Nineteen (22 %) taxa were noted as threatened, endangered, rare or decreasing on the Red List of Algae for Germany, with most at sites ≤ 26 % glacier cover, meaning further ice loss may benefit these diatoms. However, six taxa found only in rivers ≥ 28 % glacier cover may require reclassification of their Red List conservation status, as this habitat is threatened by deglaciation. Our identification of clear links between decreasing glacier cover and river diatom biodiversity suggests there could be significant reorganisation of river ecosystems with deglaciation, for example through alterations to primary production, biogeochemical cycles and the shifting resource base of alpine freshwater food webs which lack significant allochthonous energy inputs.

Introduction

Pervasive and accelerated glacier retreat in alpine regions worldwide is predicted to intensify throughout the 21st century (Intergovernmental Panel on Climate Change [IPCC], 2014). This diminishing ice cover will ultimately reduce the contribution of ice melt to rivers, subsequently increasing the relative contributions of snow melt, groundwater streams and precipitation (Brown, Hannah & Milner, 2003; Milner et al., 2017; Huss & Hock, 2018). Alteration to meltwater sourcing will physically modify channel geomorphology as diurnal and seasonal ice melt discharge peaks are attenuated, reducing erosion and reworking of proglacial sediments (Carrivick & Heckmann, 2017). Each water source also generates discrete physicochemical conditions, forming the habitat template upon aquatic communities and acting as an environmental filter to taxa which do not possess the morphological and behavioural trait combinations required to survive (Brown et al., 2018). Spatiotemporal mixing of water sources creates further diversity of habitat conditions within alpine rivers. Whilst there has been a major research focus upon the impact of glacier retreat on macroinvertebrate communities, far less is understood of how other aquatic groups will respond (Fell, Carrivick & Brown, 2017).

The sensitivity of freshwater benthic diatoms to environmental change has led to their use as representative indicator taxa in the assessment of water quality globally (Wang et al., 2014; Lobo et al., 2016). Diatom assemblages possess a diverse spectrum of ecological optima and tolerances and they reassemble in response to alterations in physicochemical environment, which underpins their use in assessment of the condition of freshwater ecosystems required by the European Water Framework Directive (Kelly et al., 2008; Lobo et al., 2016). However, there remains a clear need for knowledge of diatom assemblage responses to natural and indirect anthropogenic change, such as glacier retreat.

Benthic diatoms, alongside other biofilm components including cyanobacteria and other algae, play a major role in primary production within alpine rivers (Rott et al., 2006; Battin et al., 2016). This autochthonous input is critical to these above-treeline systems which receive minimal energy subsidy from the riparian zone (Zah & Uehlinger, 2001). Alpine rivers, particularly springs, have been identified as potential hotspots of benthic diatom biodiversity, hosting rare and threatened taxa, often in high abundance (Rott et al., 2006; Cantonati et al., 2012). Hannaea arcus and the genera Achnanthidium, Fragilaria and Odontidium are consistently the most abundant taxa within diatom assemblages across the European Alps, Himalaya and Rocky Mountains, with new species belonging to the latter recently identified in mountain streams (Hieber et al., 2001; Gesierich & Rott, 2012; Nautiyal, Mishra & Verma, 2015; Jüttner et al., 2015, 2017). However, a more complete consideration of benthic diatoms is needed to inform understanding of alpine river biodiversity responses to glacier retreat, given their role as a principal food source for invertebrate primary consumers in glacier-fed rivers (Clitherow, Carrivick & Brown, 2013).

Previous research investigating river diatom assemblages within mountain catchments has considered glacial influence with regard to distance from ice margins (Nautiyal, Mishra & Verma, 2015) and water source origins (Hieber et al., 2001). However, holistic predictions of aquatic community response to future glacier retreat require approaches that identify alterations to alpine freshwater biodiversity along a quantified spectrum of glacial influence (Brown, Hannah & Milner, 2007). Whilst such chronosequence approaches have been used recently to determine the response of macroinvertebrate, algae (excluding diatoms) and microbial prokaryote communities to deglaciation (Rott et al., 2006; Brown, Hannah & Milner, 2007; Ren et al., 2017), they are yet to be applied to benthic diatom assemblages. The efficacy of alpine freshwater conservation strategy is critically dependent upon understanding these responses, particularly for taxa that are vulnerable to extirpation due to limited motility and dispersal capacity (Liu et al., 2013).

In this study, we examined diatom assemblage structure and the abundance of individual species in rivers draining the eastern European Alps. Although proglacial regions of the Alps host high aquatic alpine biodiversity, glaciers are in long-term retreat, with approximately two-thirds of total glacier volume lost since 1850 (Zemp et al., 2006), and a further 4 to 18 % reduction of the 2003 ice area predicted by 2100 (Huss, 2012). This study utilised a chronosequence approach, sampling river sites within watersheds hosting different percentages of permanent ice cover, to provide a gradient of catchment glacier cover and in turn, a proxy for the stages of glacier retreat. This study aimed to (i) quantify the biodiversity of diatom assemblages present in alpine rivers along the catchment glacier cover gradient, (ii) determine taxon level responses to glacier cover and (iii) investigate the environmental drivers underpinning glacial influence upon alpine river benthic diatoms. Our research design facilitated novel investigation of diatom assemblage response to decreases in glacier cover within the European Alps.

Materials and Methods

Study area

Field observations were made in the central Austrian Alps (Figure 1) during June/July 2015 and 2016. Variability of glacier coverage within a small geographical region provided a broad spectrum of glacier cover whilst minimising large-scale differences due to climate or other catchment characteristics (Robson et al., 2016). Thirteen river sites spanning a gradient of catchment glacier cover (0 to 64 % permanent ice cover) were identified throughout the Eisboden, Obersulzbach and Rotmoos valleys (Table 1). Sites were selected with minimal direct anthropogenic influence and where sampling was possible at locations above the treeline. Sites included streams predominantly fed by glacial meltwaters (> 50 % catchment glacier cover) (O1, R1), mixed streams draining melt, springs and aquifer upwelling (25 – 50 % catchment glacier cover) (E2, E3, O3, R2, R3, R4, U1) and those sourced predominantly (1 – 25 % catchment glacier cover) (E1, U2) and entirely by groundwater flow (0 % catchment glacier cover) (E4, O2), to represent varying glacier cover (Table 1).

Permanent ice cover of discrete catchments pertaining to individual river sampling sites was determined using the watershed analysis function of ArcMap™ 10.4. Flow direction and accumulation models were applied to a filled 10 m ASTER Digital Elevation Model (DEM) (GitHub, 2016), to determine downslope water flow direction based upon local topography. Automated watershed delineation followed river network boundaries and were then checked manually using high resolution aerial photography, to avoid error induced by DEM resolution and to achieve the most accurate representation of local geomorphology. Regional ice extents (Glaciology Commission, 2015) were constrained to watershed boundaries to calculate the percentage of catchment area covered by ice (Table 1).

Field sampling

Water temperature, electrical conductivity (EC) and pH were measured in-situ at each site using Hanna Instruments (Woonsocket, RI, USA) H19063, HI9033 and HI98130 meters, respectively. Pfankuch Index bottom component estimates were collected (Pfankuch, 1975) as a proxy for channel geomorphological stability, with lower values representing more stable river-beds. Reciprocal values (1/Pfankuch Index scores) were used in statistical analyses so that larger values represented higher stability. River water (100 mL) was collected for ex-situ optical turbidity analysis using a Hanna Instruments HI93703 meter and then filtered for nutrient analysis. This included the detection of dissolved organic carbon (DOC) using an

Analytik Jena Multi N/C 2100 elemental thermal oxidation analyser, whereas phosphate (PO_4), nitrate (NO_3) and total nitrogen (TN) concentrations were analysed using a Two Skalar San ++ Continuous flow auto-analyser.

Benthic biofilms were sampled following CEN (2014). At each site, five submerged cobbles were selected randomly from riffles to represent a diversity of river site microhabitats though for sites O2 and O4, only three and four replicates were available respectively, due to sample damage during transportation (total n = 62 replicates). Benthic algae were scrubbed from a 9 cm^2 area of the upper surface of each cobble, using a plastic template and sterile toothbrush. Cobbles potentially exposed during prolonged periods of low flow were avoided. Samples were preserved within 70 % methylated spirits and stored at 4 °C prior to analysis.

Laboratory analysis

To prepare samples for microscopic analysis, organic material was removed to enable the unobstructed observation of diatom valves (CEN, 2014). The hot hydrogen peroxide (H_2O_2) method was used to reduce reaction times between H_2O_2 and organic matter. Samples were homogenised and 20 mL of 30 % H_2O_2 added to a 5 mL subsample, which was heated in a water bath at 90 °C (± 5 °C) for three hours. The remaining H_2O_2 was neutralised with 50 % hydrochloric acid (HCL) and subsamples suspended within distilled water and centrifuged at 1200 rpm for four minutes, four times. The remaining 5 mL pellet was diluted by adding 5 to 20 mL of distilled water, depending upon diatom concentration. The solutions were then homogenised with a vortex mixer (Stuart SA8) and 0.5 mL pipetted onto the centre of a 19 mm circular coverslip, which had been cleaned with ethanol. Coverslips were covered and air dried overnight. They were mounted using Naphrax®, as its high refractive index (>1.6) facilitated the clear examination of diatoms (CEN, 2014). The initial volume of each biofilm cobble sample and any dilutions were recorded and accounted for in subsequent volumetric calculations.

Diatom valves were counted and identified using light microscopy (Leica DM 2000), at ×1000 magnification (N Plan lens, 100 ×/1.25 oil PH3) in brightfield view. A minimum of 500 complete, individual diatom valves were identified to the highest resolution possible, to reflect the species composition of each replicate. For replicates with fewer than 500, all valves were counted. A single researcher performed all microscopy to prevent inter-analyst variance (Culverhouse et al., 2014). Identification followed Kelly (2000), Krammer and Lange-Bertalot (2004, 2007a, b, 2008), and Spaulding (2018) with taxonomic nomenclature following Lange-Bertalot et al., (2017). Characterisation of variation within the Achnanthidium minutissimum complex followed Potapova and Hamilton (2007).

To determine estimates of absolute diatom abundance, or valve density, the number of valves counted within coverslip transects was used to estimate the total number present upon the whole coverslip (0.5 ml) and then multiplied to per m^2 based on sample volume and rock area sampled (Scott et al., 2014). These extrapolations were averaged across rock scrub replicates for each river site, with mean valves per m^2 underpinning all diatom abundance analysis. As a minimum, half of each coverslip was screened, with transects encompassing both the coverslip centre and edges. Repeat counts of 20 % of all replicates identified an average estimated absolute abundance error of ± 3 %. This approach was adopted in preference to microsphere analysis given the significant and unpredictable variability of diatom concentrations between replicates hindering estimates of suitable microsphere to diatom ratios (Battarbee & Kneen, 1982).

Data analysis

The diatom species × abundance matrix was used to calculate summary metrics describing biodiversity at each river sampling site: (a) taxonomic richness (b) density of valves m^{-2}, (c) Pielou's evenness, (d) Shannon diversity index and (e) taxonomic richness of taxa classified on the Red List of Algae for Germany (Lange-Bertalot & Steindorf, 1996). The Red List was collated to identify taxa of algal conservation priority and despite development from research solely within German freshwaters, it was applied within this study given the absence of comparable datasets for Austrian rivers and the geographical proximity of their alpine regions. Changes to taxonomic nomenclature since Red List publication were identified following Guiry (2018). As the diatom assemblages hosted many taxa only identified at single sites in low abundance, Shannon diversity index was adopted as it does not weight common species over rare ones (Morris et al., 2014). Relationships between these indices and glacier cover were tested using general linear models (GLM) or generalised additive models (GAM) for data showing pronounced non-linear relationships, with the latter constructed using the mgcv package in R (Wood, 2011). Akaike's information criterion (AIC) values were used to determine best fit, with negative binomial and Gaussian distributions specified. Smoothing parameters for GAM were selected following the procedures outlined by Wood (2004). Model performance was evaluated using the percentage of deviance explained. GAMs and GLMs were also used to determine the relationship between catchment glacier cover and site-specific environmental parameters (mean water temperature, EC, turbidity, pH, 1/Pfankuch Index scores, nutrient concentrations).

Within-site β-diversity was calculated from the abundance of diatom valves identified in the replicate biofilm cobble scrub samples collected from individual river sites. Components of beta diversity (total dissimilarity (Sørensen), turnover (Simpson) and nestedness (Nestedness) were calculated for each river site using the betapart package of R (Baselga et al., 2017) and GLM used to describe the relationship between average beta component values and catchment glacier cover per site. Between-site β-diversity was determined by amalgamating average diatom valve abundances from replicates to determine a singular valve abundance value for each river site. Sørensen, Simpson and Nestedness indices were calculated for the species × abundance matrix. These components were related to pairwise differences (Bray-Curtis dissimilarity index) in catchment glacier cover between all river sites. Mantel tests (vegan R package) were used to determine the significance of correlation between these dissimilarity matrices.

Ordination analysis was performed to investigate diatom assemblage composition and taxon level responses along the gradient of glacier cover. Prior to analysis, the estimated average abundance (valves m^{-2}) of identified diatom taxa at each site was $\log_{10}(\times+1)$ transformed to constrain the influence of disparate records upon outcomes for data containing zero values (Khamis et al., 2014a). Non-metric multidimensional scaling (NMDS) was applied to a Bray-Curtis dissimilarity matrix using the vegan package of R (Oksanen et al., 2017). Significantly correlated ($p < 0.05$) scaled (mean = 0, SD = 1) physicochemical and nutrient vectors were fitted to the resulting configuration using the envfit procedure. Both NMDS axis scores were correlated to site-specific taxon abundances using Spearman’s rank correlation.

Results

Environmental parameters

Pfankuch Index scores (i.e. decreasing channel stability) and NO_3 concentrations increased significantly with glacier cover (Figure 2, Table 2). No other environmental parameters showed significant relationships with catchment glacier cover (Figure 2), although water temperature data collected over longer time periods from our study sites showed a negative relationship with glacier cover (Supporting Figure 1) and thus co-varied with channel stability. The range and maximum concentrations of DOC (0.3 – 3.8mg L^{-1}), PO_4 (0.001 - 0.003 mg L^{-1}), NO_3 (0.132 – 0.399 mg L^{-1}) and TN (0.066 – 0.311 mg L^{-1}) were low at all sites. There were no significant correlations between diatom taxonomic richness or valve density and these variables.

Diatom assemblages and taxon biodiversity

Diatom taxonomic richness, overall density and Shannon diversity index increased significantly as glacier cover decreased, with taxonomic richness and Shannon diversity showing pronounced thresholds of change at ~ 28 % catchment glacier cover (Figure 2). In total 85 taxa from 29 genera were identified, with taxonomic richness at individual sites ranging from 1 to 47 taxa and being greatest at lower cover (≤ 28 % cover, 32 to 47 taxa). Above 28 % glacier cover, richness did not exceed 29 taxa. Estimated average abundance spanned four orders of magnitude, from 15,111 valves m^{-2} (64 % cover), to 3.20×10^8 valves m^{-2} (0 % cover). The taxonomic richness of Red List taxa increased significantly with reducing glacier cover (Figure 2). Pielou's species evenness did not illustrate any significant trend across the gradient (Figure 2). Despite absence of relationships between catchment glacier cover and both total dissimilarity (Sørensen) and turnover (Simpson) components of between-site β-diversity, nestedness (Nestedness) was reduced significantly by ice loss (Figure 3). In contrast, nestedness (Nestedness) was the only component of within-site β-diversity to display no significant correlation with reductions in ice cover. While total dissimilarity (Sørensen) decreased, turnover (Simpson) demonstrated a unimodal response to declining catchment glacier cover (Figure 3).

The most abundant diatom species were those attributed to the A. minutissimum complex which represented 63.8 % of identified valves and were found at all sites. Other frequent (≥ 10 sites) taxa included Encyonema ventricosum (11 sites), Psammothidium helveticum and

Encyonema minutum (10 sites), generalists which were present at ten or more (77 %) sites. Many taxa occurred at just one (29 taxa) or two (10 taxa) sites, with 74 taxa (87 % of taxa) contributing less than 1 % to the estimated total abundance of sampled species. Caloneis lancettula and Eunotia trinacria were found exclusively at sites ≥ 52 % glacier cover while Chamaepinnularia mediocris, Cymbella parva, Gomphonema angustatum, Gomphonema calcareum, Meridion circulare, Reimeria sinuata f. antiqua and Stauroneis agrestis were found only within groundwater-fed streams (0 % glacier cover).

Of the 85 taxa identified, nineteen (22 %) were noted on the Red List of Algae for Germany as threatened, endangered, decreasing or rare. Navicula detenta, classified as threatened with extinction, was found at only one site. Endangered species included Achnanthidium caledonicum (9 sites), Achnanthidium trinodis (1 site), Encyonema hebridicum (1 site), Encyonema neogracile (4 sites), Fragilariforma constricta (1 site), Staurosira construens (1 site), Rossithidium petersenii (5 sites) and Staurosirella lapponica (2 sites). Red List species were found in greatest abundance (representing 4.3 to 24.3 % of individuals) at river sites ≤ 26 % glacier cover (Figure 2), with A. caledonicum the most abundant, representing 6.1 % of identified individuals. Sites with more glacier cover had a lower number of these species (Figure 2) and of the six taxa found exclusively in river sites with > 28 % glacier cover, only two (F. constricta, S. construens) are currently noted on the Red List.

A Shepard Plot ($r^2 = 0.99$) for the NMDS ordination (stress = 0.023) indicated the accurate preservation of rank orders within the two-dimensional display (Figure 4), ensuring that the resulting NMDS configuration represented the original distribution of data within the dissimilarity matrix as closely as possible. The NMDS Axis 1 was significantly ($p < 0.05$) correlated to the density of 23 diatom taxa (Table 3). The strongest positive correlations were between Axis 1 and A. minutissimum complex species (A. lineare, A. minutissimum, A. caledonicum, A. minutissimum var. cryptophala). No abundances correlated significantly to

NMDS Axis 2. Fitting of significantly correlated environmental parameters illustrated the strong association of glacier cover ($r^2 = 0.55$, $p = 0.013$) and 1/Pfankuch Index scores ($r^2 = 0.79$, $p = 0.001$) with Axis 1, whilst pH ($r^2 = 0.44$, $p = 0.039$) and NO_3 concentration ($r^2 = 0.39$, $p = 0.078$) were more closely aligned to Axis 2 (Figure 4).

Discussion

This study provides new insights into the response of alpine river benthic diatom assemblages to decreasing catchment glacier cover. The impact of glacier retreat upon microalgae remains poorly quantified in comparison to other aquatic groups (Rott et al., 2006; Fell, Carrivick & Brown, 2017) but this study has contributed three original findings. First, alpine river benthic diatom biodiversity and individual taxon densities were influenced significantly by reducing catchment glacier cover. Second, reductions in glacier cover will increase α-diversity but reduce β-diversity, with many taxa potentially becoming threatened or rare. Third, our research predicts some diatom taxa will be winners (i.e. expanded habitat availability) but others losers in response to glacier retreat, implying a need to reclassify the conservation status of many Austrian alpine river diatom taxa.

Environmental parameters

Pfankuch Index scores and NO_3 concentrations were the only measured environmental parameters significantly influenced by catchment glacier cover, although longer-term water temperature measurements collected outside of the sampling period suggest thermal drivers are also likely to be important. This finding is reinforced by long-standing ideas about water temperature and channel stability being key drivers of alpine river macroinvertebrate communities (Milner & Petts, 1994; Milner et al., 2001; Brown, Milner & Hannah 2007; Cauvy-Fraunié et al., 2015; Lencioni, 2018). The response of alpine benthic diatom assemblages to reducing catchment glacier cover may be driven by subsequent increases in channel geomorphological stability due to lower spatiotemporal discharge variability and river-bed movement (Biggs, Stevenson & Lowe, 1998; Carrivick & Heckmann, 2017). Benthic diatom taxa can be resilient to high flow velocities, particularly those possessing streamlined forms, low motility and strong attachment to benthic substrates (Hieber et al., 2001); morphological traits expressed by Achnanthidium spp., H. arcus and Fragilaria spp. identified at many river sites. However, the shear stress, abrasion and scouring induced by sustained or repeated channel destabilisation resets diatom assemblage succession and restricts taxonomic richness and density (Wellnitz & Rader, 2003; Bona, La Morgia & Falasco, 2012). A reduction in channel reconfiguration events linked to decreasing glacier cover may limit the abundance of generalist pioneer taxa whilst favouring species associated with later stages of succession (Biggs, Stevenson & Lowe, 1998; Kelly et al., 2008). NO_3 concentrations were significantly elevated at higher glacier cover, reflecting snow pack and sub-glacial sources and processing of N compounds (Wynn et al., 2007), but declined with glacier cover. While there was no significant correlation between taxonomic richness or valve density and NO_3 concentrations, elevated diatom densities at lower glacier cover sites with low NO_3 concentrations might indicate more efficient uptake of nutrients into biofilms as glaciers are lost.

In addition to glacier runoff, precipitation and snowmelt induced peak flows can destabilise river channels. As such, the low outlying taxonomic richness and diatom density values for the Rotmoosache (Figure 2k,l; catchment glacier cover of 30 %, 38 %, 41 %, 64 %) may have been influenced by a high flow event (discharge ~ 3 times the month's pre-flood mean) three days prior to sampling. This event would likely have contributed to sediment mobility and biofilm displacement, putting further constraints on diatom community composition alongside the effects of glacier runoff, prior to the attainment of peak flow. This finding suggests the influence of local weather conditions in combination with catchment scale deglaciation patterns upon diatom assemblage structure (Hannah et al., 2007).

Diatom assemblages and biodiversity

Benthic diatom richness increased significantly with reducing catchment glacier cover. This finding is supported by previous studies based upon sampling distance from glacier margins in the Canadian Rockies (Gesierich & Rott, 2012) and comparison of alpine rivers fed by different water sources (glacier, snowmelt, lake) in the Swiss Alps (Hieber et al., 2001; Robinson & Kawecka, 2005). In our study, high catchment glacier cover significantly constrained absolute abundance with total valve density at the most glacial site (~75 × 10^3 valves m^{-2}) comparable to maximum densities identified adjacent to the Ödenwinkelkees terminus (~81 × 10^3 valves m^{-2}) by Clitherow, Carrivick and Brown (2013). We found reductions in glacier cover to be associated with significantly increased diatom taxonomic richness and density, particularly < 28 % glacier cover. Increases in algal biomass were noted below 11 % glacier cover in the Andes (Cauvy-Fraunié et al., 2016) and Jacobsen et al. (2012) also identified the highest α-diversity for macroinvertebrates to occur at low catchment glacier cover (5 – 30 %). While the rate of increase in diatom taxonomic richness reduced below 3 % glacier cover in our study, the unimodal response observed for macroinvertebrates by Jacobsen et al. (2012) was not identified and complete deglaciation resulted in the highest diatom α-diversity.

Shannon diversity was reduced significantly following a peak at 28 % glacier cover, further indicating this threshold in diatom responses to ice loss. The harsh physicochemical conditions of sites with a high percentage of catchment glacier cover have been identified to reduce the diversity of macroinvertebrates (Brown et al., 2015), bacteria (Freimann et al., 2013), microbes (Wilhelm et al., 2013), nematodes and rotifers (Eisendle-Flöckner et al.,

2013) and other diatom assemblages (Thies et al., 2013). It is important to note that the percentage glacier cover of many catchments in the Austrian Alps has already declined below 28 % cover (Koboltschnig & Schöner, 2011) and changes in biodiversity of alpine river diatom assemblages may already have begun.

The unimodal response of within-site β-diversity (Simpson) to diminishing catchment glacier cover demonstrated increased turnover at mid-levels of ice cover, suggesting elevated patchiness of diatom habitat conditions, potentially driven by the greater co-existence of grazing and competing species at these intermediate levels of physicochemical disturbance (Roxburgh, Shea & Wilson, 2004; Khamis et al., 2016). Deglaciation and subsequent reductions in within-site β-diversity (total dissimilarity, turnover) may result from stronger biotic pressures upon diatoms (competition from other biofilm components, grazing, parasitism) within groundwater dominated flows (Khamis et al., 2016). This, coupled with reductions in the variability and magnitude of meltwater discharge pulses and more stable river beds at low glacier cover, will reduce the patchy occurrence of diatom taxa within riffle complexes, homogenising river habitats and limiting within-site β-diversity (turnover).

Changes to between-site β-diversity were driven primarily by reductions in nestedness along the gradient of glacier cover, potentially due to the absence of vulnerable cold stenothermic species or presence of groundwater-fed stream specialists assembling in rivers with lower glacier cover (Brown, Milner & Hannah, 2007). As diatom assemblages at sites with high catchment glacier cover appear to be comprised of taxa also found within other sampled river sites, future loss of glacier cover will further reduce β-diversity both within and between-sites. This response has also been identified for invertebrates (Jacobsen et al., 2012) and bacterial communities within alpine rivers, as alterations to water sourcing associated with declining glacier cover reduces habitat variability at a landscape scale (Freimann et al., 2013; Wilhelm et al., 2013). Aquatic groups therefore appear to express uniformity of response to the homogenisation of river habitats induced by glacier retreat, suggesting they are responding in common to physicochemical drivers and/or having strong inter-linkages via food webs.

The rivers sampled in this study were characterised by a small number of highly abundant generalist diatom species found at many sites, and a larger number of specialist species occurring exclusively at a few sites, often in low densities. The most abundant generalist taxa were those representing the A. minutissimum complex, which are noted for their cosmopolitan distribution along water temperature, nutrient and pH gradients, within alpine and temperate river systems globally (Potapova & Hamilton, 2007; Kelly et al., 2008). Previous research has documented the high abundance of oligotrophic, cold adapted Achnanthidium spp., Odontidium mesodon and H. arcus within catchments in the Alps (Gesierich & Rott, 2004), Himalaya (Cantonati et al., 2001) and Rocky Mountains (Gesierich & Rott, 2012). Of these, Achnanthidium spp. (particularly A. minutissimum) had a low score on Axis 1 of the NMDS ordination suggesting that these were primary colonisers in situations where channel stability was low. By contrast, species with high scores on Axis 1 (i.e. higher channel stability) tended to display a wider range of growth forms including chain-formers (e.g. O. mesodon, Staurosirella pinnata) and motile species (Nitzschia soratensis) that, taken together, suggest more mature biofilms subject to less disturbance and increased competition (Biggs, Stevenson & Lowe, 1998).

Two species (C. lancettula, E. trinacria) were identified exclusively at river sites ≥ 52 % glacier cover. Although these taxa are not documented as cold stenothermic species and display a diversity of habitat preferences in non-alpine catchments, their low motility may drive geographical and genetic isolation within fragmented deglaciating watersheds, despite their tolerance of local habitat conditions (Liu et al., 2013, Dong et al., 2016; Lange-Bertalot et al., 2017). Nineteen taxa were defined on the Red List as threatened, endangered, decreasing or rare (Lange-Bertalot & Steindorf, 1996). Achnanthidium caledonicum, listed as endangered, was identified in relatively high abundance at nine sites, suggesting that Austrian alpine rivers may act as a refuge for this species. Previous attention has been drawn to the regional importance of alpine springs as habitats of high freshwater diversity (Cantonati et al., 2012), but the occurrence of A. caledonicum at sites influenced by variable catchment glacier cover highlights the requirement to conserve a diversity of alpine rivers to protect rare benthic diatom taxa.

In contrast to threatened taxa, glacier retreat could lead to habitat expansion for some species, as reduced meltwater inputs lead to greater groundwater contributions in upstream reaches, opening up possibilities for range expansion of taxa which are specialised to river sites with no glacier cover, following a trend identified for macroinvertebrates (Brown, Milner & Hannah, 2007; Cauvy-Fraunié et al., 2015). For example, seven of the sampled diatom taxa stand to benefit from deglaciation and have previously been recorded in alpine rivers (e.g. S. agrestis), springs (e.g. M. circulare) (Falasco & Bona, 2011) and acidic wetland (e.g. C. mediocris, G. calcareum) (Buczkó & Wojtal, 2005), disconnected from glacier cover. Deglaciation could be particularly beneficial for populations of C. mediocris and G. calcareum which are currently noted on the Red List of Algae for Germany as having decreasing populations (Lange-Bertalot & Steindorf, 1996). Whilst maintenance of glacier cover required by some threatened diatom taxa will not be possible, other threatened taxa clearly stand to benefit from the expansion of their low glacial habitats. Alpine freshwater conservation strategies therefore need to prevent additional environmental pressures, including those imposed by water abstraction, hydroelectric power facilities, nutrient pollution and tourist infrastructure, which can impact upon both benthic diatoms and the wider aquatic biota in a variety of alpine river types (Khamis et al., 2014b).

Despite broad geographical use of the Red List for identifying the conservation status of river diatoms (Cantonati et al., 2001; Gesierich & Rott, 2004; Gesierich and Rott, 2012), application beyond its German reference sites may constrain accurate identification of localised variance in diatom abundances or characterisation of endemic taxa (Falasco & Bona, 2011). While not all sampled species were covered by the list, which has not been revised since 1996, it remains the most complete reference of potentially imperilled diatom taxa (Falasco & Bona, 2011). However, our study suggests a need for reclassification of current Red List conservation status for the six taxa found exclusively ≥ 28 % glacier cover if they are not identified in other types of river habitat, as sustained deglaciation will alter the distribution and persistence of the habitats upon which they depend (Brown, Hannah & Milner, 2007; Fell, Carrivick & Brown, 2017). Status changes may also be required for the 23 species whose abundance significantly correlated to NMDS Axis 1 (Figure 4) and in turn, to the aligned environmental vectors of catchment glacier cover and channel stability.

Wider implications of the diatom assemblage response

Overall this study has demonstrated the sensitivity of alpine benthic diatom biodiversity to reducing catchment glacier cover, a scenario predicted to continue across Austria and the wider European Alps throughout the 21st century (Zemp et al., 2006; IPCC, 2014). An important implication of this research is that alteration to diatom assemblage structure could have cascading impacts for higher trophic levels (Woodward, 2009; Clitherow, Carrivick & Brown, 2013) given their role providing energy to alpine river food webs (Rott et al., 2006) as principal primary producers and the predominant dietary component of cold-adapted macroinvertebrates (Clitherow, Carrivick & Brown, 2013). Greater numbers of studies adopting gut contents analysis for alpine macroinvertebrates are needed to determine the extent to which grazing consumers are selective feeders and the potential implications of glacier retreat and environmental warming upon capture mechanisms and feeding behaviours (Gordon et al., 2018).

The ablation rates of individual glaciers are influenced by interacting controls including catchment geomorphology and altitude, basal motion dynamics and local weather conditions, leading to spatiotemporally variable and often non-linear retreat sequences and runoff patterns within glaciated valleys and across alpine regions (Zemp et al., 2006; Huss, 2012; Robson et al., 2016). Whilst a catchment glacier cover chronosequence cannot fully capture this complexity, our research shows the approach provides a rapid, remote and effective means of quantifying glacier retreat effects on diatom communities in addition to other biotic components of river ecosystems (Rott et al., 2006; Brown, Hannah & Milner, 2007; Cauvy-Fraunié et al., 2015, 2016; Ren et al., 2017). Further research is now required in other alpine regions to determine if the identified response of benthic diatom assemblages to a shrinking cryosphere can be generalised to glacier-fed rivers globally as evidenced recently for macroinvertebrates (Brown et al., 2018).

Acknowledgements

This research was funded by a Natural Environment Research Council (NERC) scholarship (NE/L002574/1) awarded to Sarah Fell. Sarah Fell, Jonathan Carrivick and Lee Brown received additional financial support for fieldwork from the School of Geography, University of Leeds. The authors thank Rüdiger Kaufmann of the Alpine Research Centre, Obergurgl, University of Innsbruck, for the provision of laboratory facilities and river gauge information for the Rotmoosache, Austria. We also thank Joseph Mallalieu for assistance in the field and Kieran Khamis for data analysis advice.

References

Baselga, A., Orme, D., Villeger, S., Bortoli, J. De, & Leprieur, F. (2017). Betapart: Partitioning Beta Diversity into Turnover and Nestedness Components. R package version 1.4-1. http://CRAN.R-project.org/package=betapart.

Battarbee, R. W., & Kneen, M. J. (1982). The use of electronically counted microspheres in absolute diatom analysis. Limnology and Oceanography, 27(1), 184–188.

Battin, T. J., Besemer, K., Bengtsson, M. M., Romani, A. M., & Packmann, A. I. (2016). The ecology and biogeochemistry of stream biofilms. Nature Reviews Microbiology, 14(4), 251–263.

Biggs, B. J. F., Stevenson, R. J. & Lowe, R. L. (1998). A habitat matrix conceptual model for stream periphyton. Archiv fur Hydrobiologie, 143(1), 21-56.

Bona, F., La Morgia, V., & Falasco, E. (2012). Predicting river diatom removal after shear stress induced by ice melting. River Research and Applications, 28(8), 1289–1298.

Brown, L. E., Hannah, D. M., & Milner, A. M. (2003). Alpine stream habitat classification: an alternative approach incorporating the role of dynamic water source contributions. Arctic, Antarctic, and Alpine Research, 35(3), 313–322.

Brown, L. E., Hannah, D. M., & Milner, A. M. (2007). Vulnerability of alpine stream biodiversity to shrinking glaciers and snowpacks. Global Change Biology, 13(5), 958–966.

Brown, L. E., Milner, A. M., & Hannah, D. M. (2007). Groundwater influence on alpine stream ecosystems. Freshwater Biology, 52(5), 878–890.

Brown, L. E., Dickson, N. E., Carrivick, J. L., & Füreder, L. (2015). Alpine river ecosystem response to glacial and anthropogenic flow pulses. Freshwater Science, 34(4), 1201-

1215.

Brown, L. E., et al., (2018). Functional diversity and community assembly of river invertebrates show globally consistent responses to decreasing glacier cover. Nature Ecology & Evolution, 2, 325–333.

Buczkó, K., & Wojtal, A. Z. (2005). Moss inhabiting siliceous algae from Hungarian peat bogs. Studia Botanica Hungarica, 36, 21–42.

Cantonati, M., Corradini, G., Jüttner, I., & Cox, E. J. (2001). Diatom assemblages in high mountain streams of the Alps and the Himalaya. Nova Hedwigia, 12, 37–61.

Cantonati, M., Füreder, L., Gerecke, R., Jüttner, I., & Cox, E. J. (2012). Crenic habitats, hotspots for freshwater biodiversity conservation: toward an understanding of their ecology. Freshwater Science, 31(2), 463–480.

Carrivick, J. L., & Heckmann, T. (2017). Short-term geomorphological evolution of proglacial systems. Geomorphology, 287, 3-28.

Cauvy-Fraunié, S., Espinosa, R., Andino, P., Jacobsen, D., & Dangles, O. (2015). Invertebrate metacommunity structure and dynamics in an Andean glacial stream network facing climate change. PLoS ONE, 10(8).

Cauvy-Fraunié, S., Andino, P., Espinosa, R., Calvez, R., Jacobsen, D., & Dangles, O. (2016). Ecological responses to experimental glacier-runoff reduction in alpine rivers. Nature Communications, 7, 1–7.

Clitherow, L. R., Carrivick, J. L., & Brown, L. E. (2013). Food web structure in a harsh glacier-fed river. PLoS ONE, 8(4).

Comité Européen de Normalisation (CEN) (2014). Water Quality - Guidance standard for the routine sampling and preparation of benthic diatoms from rivers and lakes. EN

13946. Comité Européen de Normalisation, Geneva.

Contribution to the Fifth Assessment Report of the Intergovernmental Panel on Climate Change (IPCC). (2014). Climate Change 2013 - The Physical Science Basis, 1542, 1–30.

Culverhouse, P. F., Macleod, N., Williams, R., Benfield, M. C., Lopes, R. M., & Picheral, M. (2014). An empirical assessment of the consistency of taxonomic identifications. Marine Biology Research, 10(1), 73–84.

Dong, X. et al., (2016). Flow directionality, mountain barriers and functional traits determine diatom metacommunity structuring of high mountain streams. Scientific Reports, 6.

Eisendle-Flöckner, U., Jersabek, C. D., Kirchmair, M., Hashold, K., & Traunspurger, W. (2013). Community patterns of the small riverine benthos within and between two contrasting glacier catchments. Ecology and Evolution, 3(9), 2832–2844.

Falasco, E., & Bona, F. (2011). Diatom community biodiversity in an alpine protected area: A study in the maritime Alps natural park. Journal of Limnology, 70(2), 157–167.

Fell, S. C., Carrivick, J. L., & Brown, L. E. (2017). The multitrophic effects of climate change and glacier retreat in mountain rivers. BioScience, 67(10), 897–911.

Freimann, R., Bürgmann, H., Findlay, S. E. G., & Robinson, C. T. (2013). Bacterial structures and ecosystem functions in glaciated floodplains: Contemporary states and potential future shifts. ISME Journal, 7(12), 2361–2373.

Gesierich, D., & Rott, E. (2004). Benthic algae and mosses from aquatic habitats in the catchment of a glacial stream (Rotmoos, Ötztal, Austria). Berichte Des Naturwissenschaftlichen-Medizinischen Verein Innsbruck, 91, 7–42.

Gesierich, D., & Rott, E. (2012). Is diatom richness responding to catchment glaciation? A case study from Canadian headwater streams. Journal of Limnology, 71(1).

GitHub. (2016). GitHub. Retrieved May 4, 2016, from https://github.com/openterrain/openterrain/wiki/Terrain-Data

Glaciology Commission. (2015). Austrian Glacier Inventory. Retrieved May 4, 2016, from http://www.glaziologie.at/gletscherinventar.html

Gordon, T. A. C., Neto-Cerejeira, J., Furey, P. C., & O’Gorman, E. J. (2018). Changes in feeding selectivity of freshwater invertebrates across a natural thermal gradient. Current Zoology. https://doi.org/https://doi.org/10.1093/cz/zoy011.

Guiry, M. D. (2018). Algaebase. Retrieved March 8, 2018, from http://www.algaebase.org/

Hannah, D. M., Brown, L. E., Milner, A. M., Gurnell, A. M., McGregor, G. R., Petts, G. E., Smith, B. P. G. & Snook, D. L. (2007). Integrating climate-hydrology-ecology for alpine river systems. Aquatic Conservation: Marine and Freshwater Ecosystems, 17: 636-656.

Hieber, M., Robinson, C. T., Rushforth, S. R., & Uehlinger, U. (2001). Algal communities associated with different alpine stream types. Arctic, Antarctic, and Alpine Research, 33(4), 447–456.

Huss, M. (2012). Extrapolating glacier mass balance to the mountain-range scale: The European Alps 1900-2100. Cryosphere, 6(4), 713–727.

Huss, M., & Hock, R. (2018). Global-scale hydrological response to future glacier mass loss. Nature Climate Change, 8, 135–140.

Jacobsen, D., Milner, A. M., Brown, L. E., & Dangles, O. (2012). Biodiversity under threat in glacier-fed river systems. Nature Climate Change, 2(5), 361–364.

Jüttner, I., Williams, D. M., Levkov, Z., Falasco, E., Battegazzore, M., Cantonati, M., Van

De Vijver, B., Angele, C., & Ector, L. (2015). Reinvestigation of the type material Odontidium hyemale (Roth) Kützing and related species, with descriptions of four new species in the genus Odontidium (Fragilariaceae, Bacillariophyta). Phytotaxa, 234(1), 1-36.

Jüttner, I., Williams, D. M., Gurung, S., Ven De Vijver, B., Levkov, Z., Sharma, C. M., Sharma, S., & Cox, E. J. (2017). The genus Odontidium (Bacillariophyta) in the Himalaya - a preliminary account of some taxa and their distribution. Phytotaxa, 332(1), 1-21.

Kelly, M. (2000). Identification of common benthic diatoms in rivers. Field Studies, 9, 583–700.

Kelly, M.G., Juggins, S., Guthrie, R., Pritchard, S., Jamieson, B. J., Rippey, B., Hirst, H., & Yallop, M. L. (2008). Assessment of ecological status in U.K. rivers using diatoms. Freshwater Biology, 53(2): 403-422.

Khamis, K., Hannah, D. M., Brown, L. E., Tiberti, R., & Milner, A. M. (2014a). The use of invertebrates as indicators of environmental change in alpine rivers and lakes. Science of the Total Environment, 493, 1242–1254.

Khamis, K., Hannah, D. M., Clarvis, M. H., Brown, L. E., Castella, E., & Milner, A. M. (2014b). Alpine aquatic ecosystem conservation policy in a changing climate. Environmental Science and Policy, 43, 39–55.

Khamis, K., Brown, L. E., Hannah, D. M., & Milner, A. M. (2016). Glacier-groundwater stress gradients control alpine river biodiversity. Ecohydrology, 9: 1263-1275.

Koboltschnig, G. R., & Schöner, W. (2011). The relevance of glacier melt in the water cycle of the Alps: The example of Austria. Hydrology and Earth System Sciences, 15(6), 2039–2048.

Krammer, K., & Lange-Bertalot, H. (2004). Bacillariophyceae: 4. Teil: Achnanthaceae, Kritische Ergänzungen zu Achnanthes s.l., Navicula s.str., Gomphonema. In: H. Ettl, G. Gärtner, H. Heynig, D. Mollenhauer (Eds.). *Süßwasserflora von Mitteleuropa.* 2/4. Elsevier, München.

Krammer, K., & Lange-Bertalot, H. (2007a). Bacillariophyceae: 1. Teil: Naviculaceae. In: H. Ettl, J. Gerloff, H. Heynig, D. Mollenhauer (Eds.). *Süßwasserflora von Mitteleuropa.* 2/1. Elsevier, München.

Krammer, K., & Lange-Bertalot, H. (2007b). Bacillariophyceae: 2. Teil: Bacillariaceae, Epithemiaceae, Surirellaceae. In: H. Ettl, J. Gerloff, H. Heynig, D. Mollenhauer (Eds.). *Süßwasserflora von Mitteleuropa.* 2/2. Elsevier, München.

Krammer, K., & Lange-Bertalot, H. (2008). Bacillariophyceae: 3. Teil: Centrales, Fragilariaceae, Eunotiaceae. In: H. Ettl, J. Gerloff, D. Mollenhauer (Eds.). *Süßwasserflora von Mitteleuropa.* 2/3. Springer, Heidelberg.

Lange-Bertalot, H., & Steindorf, A. (1996). Rote liste der limnischen kieselalgen (Bacillariophyceae) Deutschlands. Schriftenreihe Fur Vegetationskunde, 28, 633–677.

Lange-Bertalot, H., Hofmann, G., Werum, M., & Cantonati, M. (2017). Freshwater benthic diatoms of Central Europe: over 800 common species used in ecological assessment. English edition with updated taxonomy and added species. (M. G. K. & H. L.-B. Edited by M. Cantonati, Ed.). Schmitten-Oberreifenberg: Koeltz Botanical Books.

Lencioni, V. (2018). Glacial influence and stream macroinvertebrate biodiversity under climate change: Lessons from the Southern Alps. Science of The Total Environment, 622-633, 563-575.

Liu, J., Soininen, J., Han, B. P., & Declerck, S. A. J. (2013). Effects of connectivity, dispersal directionality and functional traits on the metacommunity structure of river benthic diatoms. Journal of Biogeography, 40(12), 2238–2248.

Lobo, E. A., Heinrich, C. G., Schuch, M., Wetzel, C. E., & Ector, L. (2016). Diatoms as bioindicators in rivers. In: O. Necchi (Ed.), River Algae (pp. 245–271). Switzerland: Springer.

Milner, A. M., & Petts, G. E. (1994). Glacial rivers: physical habitat and ecology. Freshwater Biology, 32(2): 295-307.

Milner, A. M., Brittain, J. E., Castella, E., & Petts, G. E. (2001). Trends of macroinvertebrate community structure in glacier-fed rivers in relation to environmental conditions: A synthesis. Freshwater Biology, 46(12), 1833-1847.

Milner, A. M., et al., (2017). Glacier shrinkage driving global changes in downstream systems. Proceedings of the National Academy of Sciences, 114(37), 9770–9778.

Morris, E. K., et al., (2014). Choosing and using diversity indices: Insights for ecological applications from the German Biodiversity Exploratories. Ecology and Evolution, 4(18), 3514–3524.

Nautiyal, P., Mishra, A. S., & Verma, J. (2015). The health of benthic diatom assemblages in lower stretch of a lesser Himalayan glacier-fed river, Mandakini. Journal of Earth System Science, 124(2), 383–394.

Oksanen, J., et al., (2017). Vegan: Community Ecology Package. R Package Version. 2.4-3. http://CRAN.R-Project.org/package=vegan.

Pfankuch, D. J. (1975). Stream reach inventory and channel stability evaluation. US Department Forest Service, Northern Region, Montana.

Potapova, M., & Hamilton, P. B. (2007). Morphological and ecological variation within the Achnanthidium minutissimum (Bacillariophyceae) species complex. Journal of Phycology, 43(3), 561–575.

Ren, Z., Gao, H., Elser, J. J. & Zhao, Q. (2017). Microbial functional genes elucidate environmental drivers of biofilm metabolism in glacier-fed streams. Scientific Reports, 7: 12866.

Robinson, C., & Kawecka, B. (2005). Benthic diatoms of an alpine stream/lake network in Switzerland. Aquatic Sciences, 67(4), 492–506.

Robson, B. A., Hölbling, D., Nuth, C., Strozzi, T., & Dahl, S. O. (2016). Decadal scale changes in glacier area in the Hohe Tauern national park (Austria) determined by object-based image analysis. Remote Sensing, 8(1), 67.

Rott, E., Cantonati, M., Füreder, L., & Pfister, P. (2006). Benthic algae in high altitude streams of the Alps - A neglected component of the aquatic biota. Hydrobiologia, 562(1), 195-216.

Roxburgh, S. H., Shea, K., & Wilson, B. (2004). The intermediate disturbance hypothesis: patch dynamics and mechanisms of species coexistence. Ecology, 85(2): 359-371.

Scott, K. R., Morgan, R. M., Jones, V. J., & Cameron, N. G. (2014). The transferability of diatoms to clothing and the methods appropriate for their collection and analysis in forensic geoscience. Forensic Science International, 241, 127–137.

Spaulding, S. (2018). Diatoms of the United States. Retrieved March 13, 2018, from http://westerndiatoms.colorado.edu/

Thies, H., Nickus, U., Tolotti, M., Tessadri, R., & Krainer, K. (2013). Evidence of rock glacier melt impacts on water chemistry and diatoms in high mountain streams. Cold

Regions Science and Technology, 96, 77–85.

Wang, X., Zheng, B., Liu, L. and Li, L. (2014). Use of diatoms in river health assessment. Annual Research & Review in Biology, 4(24), 4054–4074.

Wellnitz, T., & Rader, R. B. R. (2003). Mechanisms influencing community composition and succession in mountain stream periphyton: interactions between scouring history, grazing, and irradiance. Journal of the North American Benthological Society, 22(4), 528–541.

Wilhelm, L., Singer, G. A., Fasching, C., Battin, T. J., & Besemer, K. (2013). Microbial biodiversity in glacier-fed streams. ISME Journal, 7(8), 1651–1660.

Wood, S. N. (2004). Stable and efficient multiple smoothing parameter estimation for generalized additive models. Journal of the American Statistical Association, 99(467), 673–686.

Wood, S. N. (2011). Fast stable restricted maximum liklihood and marginal liklihood estimation of semiparametric generalized linear models. Journal of the Royal Statistical Society: Series B (Statistical Methodology), 73(1), 3–36.

Woodward, G. (2009). Biodiversity, ecosystem functioning and food webs in fresh waters: Assembling the jigsaw puzzle. Freshwater Biology, 54(10), 2171–2187.

Wynn, P. M., Hodson, A. J., Heaton, T. H. E., & Chenery, S. R. (2007). Nitrate production beneath a High Arctic glacier, Svalbard. Chemical Geology, 244(1-2): 88-102.

Zah, R., & Uehlinger, U. (2001). Particulate organic matter inputs to a glacial stream ecosystem in the Swiss Alps. Freshwater Biology, 46(12), 1597–1608.

Zemp, M., Haeberli, W., Hoelzle, M., & Paul, F. (2006). Alpine glaciers to disappear within decades? Geophysical Research Letters, 33(13).

Tables

Table 1: Sampling site information.

Valley	**Sampling date**	**Aspect**	**Geology**	**Site code**	**Coordinates (N, E)**	**Altitude (m)**	**Area (km^2)**	**Catchment glacier cover (%)**
Rotmoosache	June 2016	N	Mica schists, Gneisses, Marble	R1	46.83104, 11.04022	2351	4.0	64
				R2	46.83633, 11.03612	2310	7.1	41
				R3	46.83981, 11.03206	2290	8.3	38
				R4	46.84623, 11.01827	2253	10.8	30
Obersulzbach	June 2015	NNW	Muscovite schists, Quartzites	O1	47.13371, 12.28085	1948	19.3	52
				O2	47.13319, 12.28296	1942	1.2	0
				O3	47.14214, 12.27648	1746	29.2	42
Eisboden	June/July 2015	N	Micas schists, Feldspar, Gneiss	E1	47.12436, 12.63836	2129	0.5	3
				E2	47.13125, 12.63408	2074	8.6	26
				E3	47.13413, 12.63749	2052	8.5	28
				E4	47.13477, 12.63710	2056	0.02	0
				U1	47.14075, 12.65157	2275	2.1	46
				U2	47.13979, 12.65328	2286	0.04	1

Table 2: GLM, GAM and Mantel summary statistics for the effect of glacier cover on physical, chemical and biological parameters.

Dependent variable	**Method**	**Chi-sq./ F**	**P**	**% Deviance Explained/r^2**
Physical parameters				
Area (km^2)	GLM (Gaussian)	0.33	0.91	50.0
Altitude (m)	GLM (Gaussian)	0.86	0.62	72.2
Water temperature (°C)	GLM (Gaussian)	0.55	0.59	9.9
Turbidity	GLM (Gaussian)	0.86	0.45	14.8
1/Pfankuch	GAM (Gaussian)	6.98	< 0.001	58.2
Chemical parameters				
DOC (mg L^{-1})	GLM (Gaussian)	0.97	0.36	12.2
NO_3 (mg L^{-1})	GLM (Gaussian)	14.45	0.006	67.4
TN (mg L^{-1})	GLM (Gaussian)	0.82	0.39	6.9
PO_4 (mg L^{-1})	GLM (Gaussian)	0.12	0.73	0.01
EC (µS cm^{-1})	GAM (negative binomial)	6.07	0.05	9.24
pH	GAM (negative binomial)	0.13	0.94	16.9
Biological parameters				
Taxonomic richness	GAM (negative binomial)	26.71	< 0.001	43.5
Density (valves m^{-2})	GAM (negative binomial)	481.50	< 0.001	33.5
Shannon diversity index	GAM (Gaussian)	13.77	< 0.001	73.4
Pielou's evenness	GLM (Gaussian)	4.23	0.066	22.7
Red List richness	GAM (negative binomial)	12.59	0.002	44.6
Within-*site* β-diversity				
Sørensen	GAM (Gaussian)	4.23	0.046	45.8
Simpson	GAM (Gaussian)	4.11	0.049	45.1
Nestedness	GLM (Gaussian)	0.56	0.472	4.8
Between-*site* β-diversity				
Sørensen	Mantel		0.062	0.22
Simpson	Mantel		0.997	- 0.37
Nestedness	Mantel		0.009	0.36

Table 3: Significant ($p < 0.05$) Spearman's rank correlations (rho) between valve abundance ($\log_{10}(\times + 1)$) of alpine river diatom taxa and Axis 1 of the NMDS ordination plot (Figure 4). R^2 (adj.) values of GAM/GLM relationships between valve abundance ($\log_{10}(\times + 1)$) and catchment glacier cover, 1/Pfankuch Index scores and NO_3 (mg L^{-1}) are also displayed. Taxa abbreviations correspond to Figure 4 (c). Relationships with a p-value <0.001 are highlighted (*). All Achnanthidium taxa belong to the Achnanthidium minutissimum complex.

Taxon	**Abbreviation**	**Axis 1**	**Glacier Cover (%)**	**1/ Pfankuch**	**NO_3 (mg L^{-1})**
Achnanthidium minutissimum	A. min	0.95*	0.52	0.83*	0.03
Achnanthidium lineare	A. lin	0.94*	0.63	0.56	0.00
Achnanthidium caledonicum	A. cal	0.91*	0.22	0.72*	0.00
Achnanthidium minutissimum var. cryptophala	A. cry	0.91*	0.27	0.76*	0.00
Adlafia suchlandtii	A. suc	0.72	0.65	0.42	0.19
Encyonema lange-bertalotii)	E. lb	0.71	0.54	0.41	0.03
Encyonema minutum	E. min	0.90*	0.26	0.45	0.23
Encyonema neogracile	E. neo	0.65	0.30	0.64	0.00
Encyonema silesiacum	E. sil	0.87*	0.32	0.90*	0.00
Encyonema ventricosm	E. ven	0.90*	0.36	0.46	0.00
Eunotia exigua	E. exi	0.54	0.61	0.33	0.00
Eunotia mucophila	E. muc	0.59	0.12	0.57	0.00
Fragilaria capucina	F. cap	0.63	0.10	0.65	0.00
Fragilaria cf. gracilis	F. gra	0.90*	0.37	0.88*	0.00
Gomphonema exilissimum	G. exi	0.66	0.16	0.43	0.10
Navicula cryptotenella	N. cry	0.56	0.42	0.35	0.01
Nitzschia palea var. debilis	N. deb	0.88*	0.82*	0.79*	0.11
Nitzschia soratensis	N. sor	0.87*	0.81*	0.76*	0.03
Odontidium mesodon	O. mes	0.79	0.52	0.39	0.14
Psammothidium helveticum	P. hel	0.88*	0.32	0.72*	0.00
Reimeria sinuate	R. sin	0.88*	0.32	0.55	0.00
Rossithidium petersenii	R. pet	0.77	0.21	0.42	0.06
Staurosirella pinnata	S. pin	0.86*	0.62	0.74*	0.00

Figure captions

Figure 1: Schematic diagrams illustrating the position of sampling sites within multiple glaciated valleys of the Austrian Alps. WGS 1984 World Mercator projection. Diagrams are derived from the *ArcMap™ World Imagery Basemap (Land Salzburg 07/11/15 and Land* Tyrol 08/03/15) with glacier outlines sourced from the Glaciology Commission (2015). Source credits: Esri, DigitalGlobe, GeoEye, i-cubed, USDA, USGS, AEX, Getmapping, Aerogrid, IGN, IGP, swisstopo, and the GIS Used Community.

Figure 2: Relationship between catchment glacier cover and physical ((a) watershed area (km^2), (b) site altitude (m above mean sea level), (c) river water temperature (°C), (d) optical turbidity NTU, (e) 1/ Pfankuch Index bottom components), chemical ((f) dissolved organic carbon (DOC) (mg L^{-1}), (g) nitrate (NO_3) (mg L^{-1}), (h) total nitrogen (TN) (mg L^{-1}), (i) electrical conductivity (µS cm^{-1}), (j) pH) and biological ((k) taxonomic richness, (l) density of diatom valves (valves m^{-2}*), (m) Shannon diversity, (n) Pielou's species evenness, (o)* taxonomic richness of diatoms classified as threatened, endangered, decreasing or rare on the Red List of Algae for Germany (Lange-Bertalot & Steindorf, 1996)). Nutrient data was unavailable for sites R1, R2, R3 and R4. Black lines represent statistical model and 95 % confidence intervals (dashed). Model details are provided in Table 2.

Figure 3: Relationship between catchment glacier cover and both within-*site β*-diversity, (a) – (c), and between-*site β*-diversity, (d) – *(f). Components of β*-diversity include total dissimilarity (Sørensen), turnover (Simpson) and nestedness (Nestedness). Within-site components are calculated from average Sørensen, Simpson and Nestedness values from dissimilarity matrices computed for the replicate biofilm samples collected at each river site. Between-site components are related to pairwise differences in glacier cover. For (b) and (c)

n = 12 as site R1 had a species richness of 1. Black lines represent GAM/GLM lines of best fit and 95 % confidence intervals.

Figure 4: (a) NMDS ordination plots of river sites and significantly correlated site-specific environmental vectors, (b) NMDS biplot of diatom taxa, (c), enlargement of (b) illustrating the position of taxa for which there was a significant correlation between estimated absolute abundance and NMDS Axis 1. Abbreviated taxon names are defined in Table 3.