Community structure and functioning below the streambed across contrasting geologies

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ABSTRACT

There is little understanding of the variability in the structure and function of metazoan hyporheic communities across streams draining geologies that weather to produce different pore sizes and, by extension, different hydrological conditions. In this study we selected two catchments in each of three geologies that had differing values of hydraulic conductivity and porosity, and sampled four riffles in each catchment at high and low water levels and at two depths. We found clear differences in the physical template of streams draining different geologies and in the composition and abundance of communities inhabiting the hyporheic zones of streams draining chalk/sandstone and limestone geologies. However, we did not detect any significant differences in body size, biomass or functional measures (diversity, richness, redundancy) between the geologies. Our findings imply that ecosystem functioning in streams draining geologies that produce fine grain sediments may be similar to those draining geologies that produce coarse grained sediments irrespective of differences in the physical template and community structure.

Keywords: Hyporheic zone, hyporheos, functional diversity, biomass, streambed biotope, functional redundancy
1. INTRODUCTION

The central aim of ecological science is to understand the relationship between organisms and their environment, a relationship driven by the habitat template on which evolution forges characteristic life history strategies (Southwood 1977, 1988). Determining the nature of the physical habitat template in the benthic compartment of river systems has a long history (see Townsend & Hildrew 1994). Here we focus on another connected compartment of the same system, the interface between rivers and aquifers, the ‘hyporheic zone’ (HZ), that hosts a continuous exchange of water, nutrients and materials between the surface stream and the aquifer (e.g., Knapp et al. 2017). The complex interactions of these two sources of water produce a unique and dynamic set of physicochemical conditions where there are strong gradients of pH and oxygen (e.g., Krause et al. 2009, Williams et al. 2010, Bardini et al. 2012). These gradients encourage the biogeochemical cycling of nutrients and attenuation of contaminants (e.g., Gandy et al. 2007, Lewandowski et al. 2011, Hester et al. 2013, Stegen et al. 2016). This function is mediated by microbial biofilms (e.g., Brunke & Gonser 1997) that form part of a hyporheic community that also includes small invertebrates (<1 mm in size) such as microcrustaceans, tardigrades, rotifers, small oligochaetes, chironomids and nematodes as well as the early instars of macroinvertebrates (Robertson et al. 2000, Peralta-Maraver et al. submitted). This metazoan hyporheic community (the hyporheos; Orghidan 1959) is the focus of our study.
The composition, abundance and biomass of metazoan hyporheos, and perhaps their ecological functioning, are strongly influenced by the physical and chemical conditions experienced within the HZ (Robertson & Wood 2010). Sediment grain size is an important factor because of its strong relationship to interstitial pore size, porosity and permeability (Brunke and Gonser, 1997). In turn grain size is partly dependent on the geology from which the sediments have been weathered although other factors, such as topography and land use, also play a role. Easily weathered geologies are associated with fine alluvial sediments whereas geologies resistant to weathering are associated with coarse sediments (Morrice et al., 1997). Sediment grain size determines the dimensions of the habitat available to the hyporheos and they are often most abundant in coarse-grained sediment where habitat availability is high (Strayer et al. 1997). In fine grained sediments, where pore spaces are reduced, small sized organisms such as the meiofauna are disadvantaged because there is little available habitat (Descloux et al 2014, Jones et al. 2015) whereas larger bodied invertebrates (e.g., amphipods, isopods, coleopteran Elmidae larvae) are still able to move through these sediments (Boulton et al. 2008). Thus, although no previous publications have presented data on body size across geologies with varying pore sizes, we might expect body size to be larger in rivers with small sized sediments (i.e., those draining chalk and sandstone geologies) than in rivers with larger sized sediments (draining limestone). Sediment grain size (and interstitial velocity) is also closely coupled to the hyporheic exchange rate (the movement of water between the HZ and the surface stream), which determines the supply of organic matter and dissolved oxygen and thus hyporheos abundance and distribution (Hakenkamp & Palmer 2000). Depth in the HZ is another important factor. In deeper layers the reduction in oxygen availability and pore space due to

Increased fine sediment loading (colmation), often resulting from agricultural practices, alters the streambed physical habitat template and is a major and well recognised global threat to stream ecological integrity, leading to reduced hyporheos diversity and modified community structure and functions (e.g., Jones et al. 2015, Descloux et al. 2014, Mathers et al. 2017). In contrast, there is very limited information on the underlying variability in the structure and function of metazoan HZ communities (hyporheos) across streams draining different geologies, despite the centrality of such data to understanding how fine sediment ingress will affect hyporheos.

The range of species' functional traits (functional diversity) likely underpins a key mechanistic link between species richness and ecosystem functioning and a growing body of evidence suggests that community functional diversity predicts ecosystem functioning better than species based indices (e.g. Gagic et al. 2015, Griffin et al. 2009). Functional diversity can be decomposed into functional richness and functional redundancy (Carmona et al. 2016); communities with high functional redundancy, where two ecological units have the same trait values and therefore occupy the same functional space, are thought to be more resilient to disturbances (i.e., they can lose species without a great loss of function; Carmona et al. 2016). Thus understanding how functional diversity, richness and redundancy vary across the physical habitat template in the hyporheic zone is an important goal given the
likely role of the hyporheic community in riverine ecosystem functioning. However, very few studies have used a trait-based approach for hyporheic communities and of those that have, to our knowledge, none have included measures of functional diversity, richness and redundancy (see Claret et al. 1999, Robertson & Wood 2010, Descloux et al. 2014, Bona et al. 2016, Doretto et al. 2107, Mathers et al. 2017).

The objective of this study was to assess differences in hyporheos composition and function across streams draining differing geologies. We investigated eight UK rivers of three distinct geologies with differing values of hydraulic conductivity and porosity. We sampled multiple locations incorporating upwelling and downwelling sites and two depths on two occasions. We expected that the community composition, abundance, body size, biomass, and functional measures of the hyporheos in the rivers would differ in response to the variation in these features of the physical habitat template.

We hypothesised that:

1. Hyporheos structure (in terms of composition and abundance) will vary between geologies resulting from the varying habitat template. We expect that hyporheos will be more abundant and characterised by meiofauna in geologies that weather to produce coarse-grained sediments.

2. Taxonomic richness, body size, biomass and diversity will also differ between geologies and with depth. We expect that body size will be greatest where pore size is small and the other measures will be highest where sediment size/pore space is greatest and decline with increasing depth.
3. Functional diversity, richness and redundancy will differ between geologies in response to differences in the physical habitat template. We also expect that these measures will decline with increasing depth.

2. METHODS

2.1 Survey design

Three geologies of differing hydraulic conductivity (k) and porosity (n) were studied (i) cretaceous chalk $k = 10^{-8}$-$10^{-5}$m/s, $n = 0.15 - 0.45$; (ii) permotriassic sandstone $k=10^{-10}$-$10^{-5}$m/s, $n=0.05-0.35$; (iii) carboniferous limestone $k=10^{-6}$-$10^{0}$m/s, $n= 0.05-0.5$. (Hiscock 2007). Two catchments each were selected in chalk, sandstone, limestone (unglaciated) and limestone (glaciated) geologies (Table 1). In each catchment four sites (permanently flowing riffles) were selected on the main stem and tributaries (Table 1). We ensured that sites differed as little as possible apart from geology; all sites had similar water quality, discharge, catchment area and elevation (Table 1). The land use in all catchments was predominantly pasture. All sites were located on 3rd to 4th order streams (stream order derived from Ordnance Survey maps of the local area following Strahler 1964). Stream bank widths were between 3-5 metres and streams were at least 5cm deep.

2.2. Sediment Grain Size

The assessment of sediment grain size at each site was undertaken using a variety of methods, i) broad scale site selection determined by the local geology and associated hydraulic properties of the aquifer (see above), ii) measurement of vertical hydraulic gradient (VHG) from both the head and tail of each riffle following
Baxter et al. (2003). Briefly a mini-piezometer was inserted to a depth of 30cm, purged and left to settle for 2 hours before measurement. \( \text{VHG} = (h_s - h_p)/L \). \( h_s \) is the distance the water in the mini-piezometer rises above the stream water surface. \( h_p \) is the distance from the top of the mini-piezometer to the water level in the mini-piezometer and \( L \) is the depth of the mini-piezometer in the sediment, iii) collection of sediment samples from the head of each riffle during late autumn when groundwater levels and river flow velocities were reduced. Sediment was collected using a spade (depth penetration 20cm, surface area 20cm\(^2\)), emptied into a 100 \( \mu \)m mesh net held just downstream and stored in a plastic bag. Sediment samples were air dried then weighed to the nearest gram to obtain the total sediment mass and fed into a sediment shaker following the approach of Gordon et al. (2004). Sediment fractions were expressed as a percentage of the total sediment mass. iv) at 50cm depth, granulometric conditions were assessed by recording the time taken to pump 5 litres of water because there is a strong positive relationship between hydraulic conductivity and sediment grain size (Sharp 1988; Descloux et al. 2010). The combined measures gave a weight of evidence approach to quantification of the sediment proportions and associated properties for each catchment across the three geologies.

2.3. Hyporheic community sampling:

Each riffle was sampled on two occasions 1) at high groundwater levels (summer 2008) and 2) at low groundwater levels (autumn 2008). In summer 2008, three replicate hyporheos samples were taken from the deep (50 cm) HZ at both the head (downwelling) and tail (upwelling) of each riffle (192 samples in total). In autumn 2008, the same survey design was followed except samples were additionally taken
from the shallow (20 cm) HZ (384 samples in total) because low numbers of
individuals were collected at 50 cm depth.

Samples were collected using the Bou-Rouch method (Bou and Rouch 1967; Bou
1974). Piezometers were installed at least 1 metre apart and allowed to settle for at
least one hour before pumping 5 litres of water (after Kibichi et al 2009). This was
filtered through a 63 µm sieve and preserved in 70% IMS. Preserved samples were
sorted under a stereomicroscope and photographed at calibrated magnifications.
Subsequently the length and width of all organisms (except Oligochaeta, Nematoda
and Acari) were measured to the nearest micrometre using these images and an
image analyser. If large groups of similar sized instars were found (>50 µm, primarily
Bryocamptus sp. and Chironomidae larvae) all individuals were counted, then 20%
of the group was measured to obtain a mean value which was then applied to the
whole group. The prolate ellipsoid volume of each group was calculated following
Omesova et al. (2008).

\[ V = \frac{4}{3}\pi lw^2 \]

\[ w = \text{width}, l = \text{length}, V = \text{volume (mm}^3\)\]

Invertebrates were identified to species level where possible using the following
keys: Copepoda (Einsle 1993, Janetzky et al. 1996); Cladocera (Scourfield 1994);
et al. 2003, Edington and Hildrew 2005) and Groundwater Crustacea (Gledhill 1993).
Oligochaeta, Diptera, Ostracoda and Nematoda were not identified further due to the
poor preservation of Oligochaeta and Nematoda and the predominance of early
instar Chironomidae larvae.
2.4. Water chemistry

Measurements were taken directly from each piezometer. Conductivity, dissolved oxygen (DO), temperature, and pH were determined using portable meters (pH - Hanna HI-9025; electrical conductivity - Hanna HI-9635; dissolved oxygen - temperature - Hanna HI-9145). Then, 0.1 litre of water was withdrawn from the piezometer and frozen within 12 hours. Subsequently, on the day of analysis, water samples were defrosted and passed through a 0.7 μm filter to remove particulate matter. Cations and anions were analysed using high performance anion-exchange chromatography (DIONEX ED40 electrochemical detector (minimum reporting value =1.0 mg/L; Thermo Fisher Scientific, Waltham, Massachusetts).

2.5. Functional traits

Traits were characterised for 37 taxa using trait tables from Descloux et al. (2014) for the permanent meiofauna and Tachet et al. (2010) for temporary meiofauna. Trait profiles were described mainly at genus level except for some Diptera, Oligochaeta (described as trait profiles of potential families in the corresponding biogeographic area) and Copepoda (described as Harpacticoida or Cyclopoida). Nine traits with 55 modalities incorporating a range of life history, mobility, morphological, and ecological traits were included following Descloux et al. (2014) but excluding the trait ‘fecundity’ due to lack of data (see Supplementary Information Table S25). A score was assigned to each taxon describing its affinity for that modality (Descloux et al. 2014; Tachet et al. 2010). The mean trait profile of each riffle assemblage was obtained by weighting the individual trait profiles of taxa by their total abundance in the sample (replicate and upwelling/ downwelling samples were pooled to give a single value of abundance per riffle and water level because we did not detect
significant differences in abundance depending on the direction of vertical flow).

Then, the sums of the weighted scores (one per trait category) were rescaled to sum to one for each trait and riffle and depth, following Descloux et al. (2014).

2.6. Data analysis

2.6.1 Community structure - Composition and abundance of all identified taxa during the study period was averaged by site and depth in order to merge all the recovered information of the community structure per geology treatment. Then, a NMDS analysis (Non-metric Multi-Dimensional Scaling) was performed to describe the dissimilarity among communities based on their structure (taxonomic composition and abundance). This is an effective method for the ordinations of ecological data that works with rank orders dissimilarity matrixes (rather than absolute values) and can handle nonlinear responses of the biological attributes of any shape and effectively and robustly find the underlying gradients (Oksanen, 2015; Quinn & Keough, 2002). In order to handle excessively large differences between abundance values, these were transformed using Wisconsin double standardization (Bray & Curtis, 1957). Dissimilarity matrices between communities used in the ordination were calculated with the quantitative Bray-Curtis Index. An iterative process was carried out to find the ordination with the best goodness of fit (Oksanen, 2015).

Subsequently, analysis of similarity (ANOSIM) was performed to determine whether there was a significant effect of the geology on the structure of the communities \( (p-value < 0.05) \) and the reliability of the test (ANOSIM R value). This analysis also uses a rank order dissimilarity matrix to determine whether there is a significant difference between two or more sample groups based on whether there are greater differences between these groups compared to within groups (Oksanen,
In this manner, we compared the magnitude of differences intra-treatments (ANOSIM within- and between-group rank dissimilarities are available as Supplementary information: Fig S2). Finally, post-hoc comparisons between geologies were conducted with PerMANOVA pairwise contrasts to identify those treatments that differed significantly. All analyses were performed with the Vegan package (Oksanen et al., 2013) within the R software platform (R Core Team 2014).

2.6.2 Taxonomic richness, body size, biomass, community diversity and functional measures (diversity, richness, redundancy). Taxonomic richness, body size, biomass, Shannon-Wiener diversity, functional diversity, functional richness, and functional redundancy were compared between geologies (factor with four levels: chalk, sandstone, unglaciated limestone and glaciated limestone) and depth compartment (factor with two levels: 20 and 50 cm) using low water level data and applying 2-way ANOVA tests. Then, taxonomic richness, body size, biomass, Shannon-Wiener diversity, functional diversity, functional richness and functional redundancy were also compared between geologies during the whole study period by using only data from 50 cm depth with a nested 1-way ANOVA test, correcting by water level. In order to solve heterogeneity in the residuals, body size was log transformed, but this was not necessary for the rest of responses. No differences were found in the studied responses between downwelling and up-welling conditions within riffles during data exploration. Accordingly, metrics were pooled by riffle (riffle as a replicate per site). Dependency structure of the residuals with the study site (Site) was incorporated in the previous ANOVA tests in the design of the study (random factor). In this manner, we accounted for repeated measures (non-dependence of residuals with study site in the ANOVA tests). In order to fit the
random factor to the ANOVA tests we used the \textit{lmer} function of the R package \texttt{lme4} (Bates et al. 2017, R Core Team 2016). Validation of underlying assumptions of normality and homocedasticity of tests residuals was applied following Zuur et al. (2009) (supplementary material: Fig S1). Finally, Wald chi-square tests were performed in order to assess the significance of the analysed factors on the responses using the \textit{Anova} function from the \texttt{car} R-package (Fox & Weisberg 2011) and, subsequently, post-hoc Tukey tests were applied to compare which specific treatments differ significantly using the \textit{diffsmmeans} function from the R-package \texttt{lmerTest} (Kuznetsova et al. 2015).

### 3. RESULTS

The physical habitat template showed clear differences between geologies and across water levels. Pump times were significantly higher in rivers draining chalk than in other geologies (F\textsubscript{3,176}=21.9, p<0.001, Table 2) and these findings were supported by VHG measurements (Table 2). Chalk rivers also had the highest percentage of sediments in the 0.5 – 2mm size category. Conductivity (F\textsubscript{3,563}=403.0, p<0.001), pH (F\textsubscript{3,563}=116.1, p<0.001), dissolved oxygen (F\textsubscript{3,563}=7.52, p<0.001), temperature (F\textsubscript{3,563}=200.3, p<0.001), and nitrate (F\textsubscript{3,563}=124.2, p<0.001), also differed significantly between geologies. Conductivity (F\textsubscript{1,563}=12.3, p<0.001), pH (F\textsubscript{1,563}=11.9, p<0.001), dissolved oxygen (F\textsubscript{1,563}=86.0, p<0.001), temperature (F\textsubscript{1,563}=2097.9, p<0.001), and nitrate (F\textsubscript{1,563}=4.4, p<0.001), also differed significantly between water levels. See Table 2 for means and standard errors of these parameters across geologies and water levels.
The NMDS ordination model based on taxonomic composition and abundance was run 20 times for the two-dimensional ordination with a very high goodness of fit between the distances in the ordination against the original data (stress = 0.20, linear fit \( R^2 = 0.98 \), nonmetric fit \( R^2 = 0.86 \)). Accordingly, the Shepard plot of the final model has small scatter around the fitted line (Supplementary Information Fig S1) and the bi-dimensional ordination is representative of the original differences between assemblages. The NMDS-plot clearly discriminated assemblages between geologies (Fig 1a) and showed which taxa contributed most to the observed dissimilarities (Fig 1b). ANOSIM analysis found a significant effect of geology on the observed dissimilarities between assemblages \( (R = 0.63, p\text{-value} = 0.002) \), supporting the NMDS plot. Thus our first hypothesis, community composition will differ between geologies, was upheld. The post hoc PerMANOVA pairwise contrasts detected significant differences between limestone assemblages (both glacial and un-glacial) with sandstone and chalk sites respectively. However, these differences were not significant between sandstone and chalk assemblages, or between glacial and un-glacial limestone assemblages (Fig 1a, Table S2) i.e., differences in assemblage structure (composition and abundance) between sandstone and chalk sites and between both limestones were too small to be able to characterise them as different communities. The hyporheos of Limestone catchments was characterised by a suite of stoneflies and caddisflies (e.g., *Leuctra* spp., Perlodidae, *Limnephilus* sp.) and microcrustacea (e.g., *Paracyclops* sp., *Moraria* sp., *Elaphoidella* sp.) (Fig. 1b). Chalk catchments were defined by large-bodied Crustacea (e.g., *Gammarus pulex*, *Niphargus* spp., *Crangonyx* sp.) whereas indicative taxa of sandstone catchments included harpacticoid (e.g., *Maraenobiotus* sp., *Canthocamptus* sp.) and cyclopoid (Eucyclops sp.) copepods (Fig. 1b).
Fig 1: (a) NMDS ordination model of different HZ assemblages by geology including ANOSIM statistic (R-value) and significance (p-value). Polygons group the assemblages by geology sites in the ordination. Assemblages whose structure is not significantly different after post-hoc PerMANOVA tests (P-value < 0.005) are indicated with the same letter. (b) Distribution of the most influential taxa on the ordination. In order to simplify interpretation of the plot, those taxa falling in the centre of the ordination (low influence in the ordination) have been removed. For explanations of species abbreviations see Supplementary Table 23.
Our second hypothesis; taxonomic richness, body size, biomass and Shannon-Wiener diversity will differ across geologies and depths was partly supported. Only taxonomic richness showed significant differences across some geologies with larger values in rivers draining unglaciated limestone (Fig. 2b). In contrast, depth showed a strong effect on all these responses. Taxonomic richness, body size, biomass and Shannon-Wiener diversity declined with increasing depth for all geologies (Fig. 2a, c, e, g) and in most cases these differences were significant.

3.4. Our third hypothesis; functional measures will differ with geology and decline with increasing depth was only partially upheld. Functional measures did not differ significantly across geologies either in the summer data or across the whole study period (Fig. 2i – n). However, functional richness and redundancy declined significantly with increasing depth for several geologies (Fig. 2 I,k). Numerical results and p-values from the Wald chi-square and post-hoc Tukey tests are available as Supplementary information (Tables S1 - S22).
Fig 2: Taxonomic richness, body size, biomass, Shannon-Wiener diversity and functional richness, redundancy and diversity (mean ±1SE/ 5 L). Summer data (a,c,e,g,i) are pooled across the four geologies (CC: Chalk, PTS: permo-triassic sandstone, UGL: unglaciated limestone, GL: glaciated limestone) and two depths (20cm and 50cm). While, data from 50 cm depth (b,d,f,h,j) of the whole study period are just pooled across geologies. Geologies where total responses are not significantly different are indicated with the same letter (95% CI).

4. DISCUSSION
Our study showed clear differences in the HZ physical habitat template in rivers draining different geologies at the regional scale and corresponding variations in the community composition of these HZ's. Geologies that weather easily to produce fine grained alluvial sediments (chalk and sandstone) possessed hyporheos that differed significantly from those that were present in rivers where the geology did not weather so easily resulting in coarser grained sediments (carboniferous limestones). The hyporheos in coarse-grained limestone rivers included an abundance of small-bodied copepods, which were able to exploit the larger pore spaces available. In contrast the hyporheos of chalk rivers was dominated by larger macroinvertebrates, presumably because they were able to burrow through these sediments more easily (Boulton et al. 2008). Although copepods did occur in these rivers, they were much less abundant and diverse.

We detected clear differences in taxonomic richness between geologies in agreement with previous studies (e.g. Strayer et al. 1997; Ward et al. 1998; Bo et al. 2007, Descloux et al. 2014) despite the lower % of fine sediments in our study. These authors noted that decreasing sediment size and interstitial space exert a negative effect on abundance and taxon richness (although we did not compare abundance between geologies as the taxonomic composition of the hyporheos varied so markedly). To our knowledge only one publication has compared the biomass of metazoan hyporheos across sediment types in perennial streams (Reynolds & Benke 2012). They found that the biomass of chironomid assemblages was highest in the HZ's of gravel-cobble reaches, intermediate in sand reaches and lowest in mud-silt reaches. Our study is the first to compare HZ biomass of the whole metazoan community retained on a 63μm sieve across geologies with
different pore sizes. Our finding, that biomass was similar across geologies,
suggests that the HZ’s of all these rivers support a similar quantity of hyporheos
although the structure and composition of these communities differ. Contrary to our
expectations, although rivers draining limestone included small bodied taxa and
chalk rivers were characterised by taxa with larger body sizes, this did not translate
into significant body size differences between the rivers, perhaps because
macroinvertebrate taxa were represented by very early instar individuals. Our
findings suggest that individuals of any body size are equally likely to penetrate bed
sediments of the three geologies and that the HZ’s may therefore have equal
connectivity with the surface in terms of organism movement.

Our findings, that taxonomic richness declined with increasing depth and that body
size and biomass tended to decrease with depth, concur with those of other studies
(e.g., Strayer et al. 1997, Davy-Bowker et al. 2006, Pacioglu & Robertson 2017,
Peralta-Maraver et al. submitted). This likely results from a reduction in pore space
and oxygen with increasing depth (Maridet & Philippe 1995, Strayer et al. 1997,

HZ’s are increasingly recognised as a central component of the lotic ecosystem,
particularly regarding their functional role in the attenuation of nutrients and
pollutants (e.g., Lewandowski et al. 2011; Peralta-Maraver et al. 2018) and their
importance as a refuge from surface disturbances (e.g., Robertson & Wood 2010,
Maazouzi et al. 2017). Although biodiversity is understood to be a fundamental
driver of ecosystem functioning, there is a growing acceptance that the diversity of
species functional traits – functional diversity - within a community underpins and
better describes ecosystem functioning (Gagic et al. 2015). Our study is the first to determine the functional richness, redundancy and diversity of hyporheos in contrasting geologies. Despite differences in the physical habitat template, which resulted in distinctive hyporheos per geology, none of our measures of ecosystem function significantly differed between geologies implying equal functioning across the HZ’s. Several studies have demonstrated the impact of fine sediment deposition (colmation) on individual functional traits of hyporheos (e.g., Descloux et al. 2014, Bona et al. 2016, Doretto et al. 2017, Mathers et al. 2017). We therefore expected, but did not find, that functional diversity would be lowest in the fine sediment rivers. As anticipated, we found functional measures declined with increasing depth, as did taxonomic richness suggesting that ecosystem functioning decreases with increasing depth. Thus, our results imply that maximal functioning of the metazoan hyporheos occurs in the shallow HZ in accordance with findings for microbial biofilms in the HZ (Battin et al. 2003, O’Connor & Harvey 2008, Boano et al. 2014, Knapp et al. 2017). Our findings suggest that, for these relatively unimpacted rivers draining three different geologies, the HZ’s all supported similar quantities of hyporheos, had similar connectivity with the surface stream and had similar levels of functional richness, redundancy and diversity, all of which implies relatively similar ecological functioning irrespective of differences in the physical template and community structure between geologies. However, our approach to characterising ecosystem functioning within the HZ was quite broad brush; we do not yet know the extent to which their capacity to undertake these functions is resilient to environmental perturbations i.e., the degree of response diversity within the functionally redundant elements of the community (Mori et al. 2013) and whether this differs across geologies.
5. CONCLUSION

The effects of fine sediment accumulation in streams is a global concern yet there is limited understanding of the underlying variation in hyporheos structure and function across streams draining differing geologies despite their contribution to the delivery of ecosystem services such as pollution attenuation. Such data is essential to contextualise the effect of perturbations such as fine sediment ingress. We found that despite clear differences in metazoan community structure between streams with fine sediment and streams with coarse sediment, there were no significant differences in biomass or functional measures between the geologies implying that the HZ’s in all geologies functioned equally.

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7. COMPETING INTERESTS STATEMENT

The authors have no competing interests to declare.

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organic matter, and sediment granulometry as controls on hyporheic animal


Table 1. Physical and water quality characteristics of the studied catchments

<table>
<thead>
<tr>
<th>Geology</th>
<th>Site location</th>
<th>Catchment area (Km²)</th>
<th>Naturalized mean annual discharge (m³S⁻¹)</th>
<th>Elevation (m)</th>
<th>RBMP C2 status</th>
<th>MI²</th>
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<tbody>
<tr>
<td>Chalk</td>
<td>RIVER FROME</td>
<td>30-40</td>
<td>0.31-1.25</td>
<td>78-95</td>
<td>High</td>
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<td></td>
<td>N50° 47' 23.0&quot; W002° 34' 49.9&quot;</td>
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<td>N50° 46' 36.3&quot; W002° 28' 10.4&quot;</td>
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<td>N50° 45' 07.3&quot; W002° 30' 42.6&quot;</td>
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<td>N53° 00' 00.7&quot; W001° 47' 30.0&quot;</td>
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1 Data source [http://environment.data.gov.uk/catchment-planning/](http://environment.data.gov.uk/catchment-planning/)

Table 2: Means (±1 SE) of selected physico-chemical parameters for three geologies and two water levels. Pump time = time (in minutes) to pump 5 litres of water.

<table>
<thead>
<tr>
<th>Geology</th>
<th>Vertical Hydraulic Gradient. H = riffle head, T = riffle tail</th>
<th>Water level</th>
<th>% sediment (0.5-2mm)</th>
<th>% sediment (4-16mm)</th>
<th>Pump time</th>
<th>Conductivity (µS)</th>
<th>pH</th>
<th>Dissolved oxygen (mg/L)</th>
<th>Temperature (°C)</th>
<th>NO₃ (mg/L)</th>
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<tbody>
<tr>
<td>Chalk</td>
<td>0.008±0.003 (H)</td>
<td>High</td>
<td>30</td>
<td>48</td>
<td>542.2±7.39</td>
<td>7.7±0.03</td>
<td>5.8±0.17</td>
<td>18.2±0.15</td>
<td>17.2±0.75</td>
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<tr>
<td></td>
<td>0.0006±0.002 (T)</td>
<td>Low</td>
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<td></td>
<td>2.9±0.2</td>
<td>560.7±5.03</td>
<td>7.5±0.03</td>
<td>7.9±0.21</td>
<td>12.3±0.09</td>
<td>14.3±0.80</td>
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<td>Sandstone</td>
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<td>High</td>
<td>17.5</td>
<td>62.5</td>
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<td>15.2±1.14</td>
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<td>0.03±0.005 (T)</td>
<td>Low</td>
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<td>1.6±0.1</td>
<td>317.2±11.5</td>
<td>7.3±0.04</td>
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<td>6.1±0.19</td>
<td>15.0±0.1</td>
<td>1.3±0.07</td>
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<td>0.08±0.011 (T)</td>
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<td>1.5±0.1</td>
<td>243.9±9.2</td>
<td>7.8±0.02</td>
<td>7.7±0.18</td>
<td>10.6±0.07</td>
<td>1.5±0.07</td>
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<tr>
<td>Limestone</td>
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