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DOCTOR OF PHILOSOPHY

Quantifying the response of macroinvertebrates to gradients of fine sediment pollution

McKenzie, Morwenna

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# Quantifying the response of macroinvertebrates to gradients of fine sediment pollution

By

Morwenna Mckenzie

PhD

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A thesis submitted in partial fulfilment of the University's requirements for the Degree of Doctor of Philosophy



#### **Certificate of Ethical Approval**

Applicant:

Morwenna McKenzie

Project Title:

Quantifying the response of macroinvertebrates to gradients of fine sediment pollution

This is to certify that the above named applicant has completed the Coventry University Ethical Approval process and their project has been confirmed and approved as Medium Risk

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### Abstract

Erosion, transportation and deposition of fine sediment (organic and inorganic particles <2 mm in diameter) are fundamental processes in the hydrogeomorphic cycle and river systems require a constant supply in order to function. However, excessive fine sediment delivery can cause serious deleterious effects to aquatic systems and is one of the leading causes for failure to meet Good Ecological Status as set out by the EU Water Framework Directive (2000/60/EC). Given the need for effective management of fine sediment, this thesis examines how fine sediment is driving macroinvertebrate responses in order to help improve biomonitoring, i.e. the practice of using biological communities to track environmental change. A systematic style review was undertaken to assess the weight of evidence for macroinvertebrate responses to fine sediment, which identified several correlative relationships. However, a global imbalance of evidence is apparent and there is a distinct knowledge gap of the mechanisms driving macroinvertebrate responses to fine sediment. The review outcomes helped inform the design of a controlled laboratory experiment which investigated the direct physical effects of fine sediment (e.g. clogging and abrasion of gills) on three different species of macroinvertebrates. The results showed that gill surfaces were covered in fine sediment debris to varying extents and responded differently to treatments in a way that suggested gill morphology and behavioural responses (such as avoidance) as key factors.

The last decade has seen a development in sediment-specific biomonitoring tools globally. Through a national (England) fieldwork sampling regime, existing sediment-specific biomonitoring indices were tested against varying gradients of fine sediment (deposited and suspended) alongside indices for general ecological health. Further insights into the response of macroinvertebrates (both taxonomic and trait-based) to fine sediment were explored using a variety of statistical techniques. The results reinforced several outcomes of the earlier systematic style review and also supported the use of sediment-specific biomonitoring indices. However, the majority of variation in sediment-specific

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index scores at each site were related to habitat and flow variables. Finally, the results obtained within this thesis were linked with emerging ecological theory and the factors which may influence the success of biomonitoring indices globally (e.g. invasive non-native species and climate change). This thesis ends by making recommendations for monitoring approaches and future research directions.

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## List of abbreviations

AIC	Akaike's Information Criterion
ASL	Agricultural Sediment Loading
ASPT	Average Score Per Taxon (often used when referring to the
	WHPT index)
ASR	Agricultural Sediment Risk rating
BMWP	Biological Monitoring Working Party (biotic index)
CoFSI	Combined Fine Sediment Index
CWM	Community Weighted Means
EA	Environment Agency
EA	Environment Agency
EPSI	Empirical Proportion of sediment Sensitive Invertebrates
EPSImixed	Empirical Proportion of sediment Sensitive Invertebrates
	including mixed taxon level scoring
EU	European Union
FD	Functional Diversity
FDis	Functional Dispersion
FRic	Functional Richness
FSSR	Fine Sediment Sensitivity Risk (using in PSI/EPSI)
GF	Gradient Forest
GIS	Geographic Information System
IMS	Industrial Methylated Spirits
INNS	Invasive Non-Native Species
LIFE	Lotic Index for Flow Evaluation
LOI	Loss on Ignition
NRMSE	Normalized Root Mean Square Error
NTAXA	Number of Taxa (often used when referring to the WHPT
	index)
NTU	Nephelometric Turbidity Units
oFSI	Organic Fine Sediment Index (constituent of the CoFSI)

PC	Principal Component
РСА	Principal Components Analysis
PICO	Population, Intervention/Exposure, Comparator and Outcome
	(used for developing evidence review scope)
PSI	Proportion of sediment Sensitive Invertebrates
REA	Rapid Evidence Assessment
RHS	River Habitat Survey
RICT	River Invertebrate Classification Tool
RIVPACS	River Invertebrate Prediction and Classification System
SD	Standard Deviation
SEM	Scanning Electron Microscope
SM	Systematic Map
SR	Systematic Review
SSC	Suspended Sediment Concentration
TITAN	Threshold Indicator TAxa ANalysis
ToFSI	Total Fine Sediment Index (constituent of the CoFSI)
UKTAG	United Kingdom Technical Advisory Group
VIF	Variance Inflation Factor
WFD	Water Framework Directive (EU)
WHPT	Whalley-Hawkes-Paisley-Trigg index

## **Chapter 1 – Introduction**

#### **1.1 The research context**

Freshwater environments represent a unique link between terrestrial and climatic systems. These environments provide important roles other than the provision of drinking water, including ecosystem services such as flood retention, water purification, nutrient cycling, groundwater recharge, biodiversity, tourism, and recreation (Wilson and Carpenter 1999; Arthington et al. 2010). These systems therefore have high natural capital value, contributing to the estimated \$125 trillion total value of ecosystem services per year (Costanza et al. 2014)<sup>1</sup>. Lakes and rivers alone have an estimated value of over \$2.5 trillion per annum (Costanza et al. 2014)<sup>2</sup>.

Loeb and Spacie (1994, p3) stated that '*the health of an aquatic ecosystem is degraded when the ecosystem's ability to absorb a stress has been exceeded'*. Stress on an ecosystem can be physical, chemical or biological but these stressors are not mutually exclusive and can occur in combination leading to faster, and possibly prolonged, environmental degradation. Humankind's reliance on aquatic systems contributes to their susceptibility to environmental degradation from anthropogenic stressors. This is further exacerbated as human activities have increased the demand on environmental resources and the ecosystem services that they provide (Vörösmarty et al. 2010). Fine sediment, defined as organic and inorganic particles <2 mm in diameter, in aquatic environments has been recognised as a significant problem for over 40 years when it was first described as the most detrimental aquatic pollutant (Ritchie 1972). Increasingly intensive agricultural land management, construction, mining, deforestation, and in-channel modifications leading to bank erosion and channel incision, are some of the main anthropogenic sources

<sup>&</sup>lt;sup>1</sup> Equivalent to £74.2 trillion as per the GBP exchange rate on 20 May 2014 (date of publication of Costanza et al. 2014)

<sup>&</sup>lt;sup>2</sup> Equivalent to £1.4 trillion (calculated as per above footnote)

leading to increased sediment loads in rivers (Owens et al. 2005, Collins et al. 2009a, Yule, Boyero, and Marchant 2010). The effects on aquatic environments are severe, including flooding, navigation blockages, and wide-ranging impacts on biota. There is an urgent requirement for targeted monitoring to determine where management methods are required to reduce the delivery of fine sediment to aquatic environments.

The configuration of aquatic health monitoring in the UK is currently bound by its obligations to the European Union (EU). The EU Water Framework Directive (WFD) (European Community 2000/60/EC) requires all ground and surface water bodies (rivers, lakes, transitional and coastal waters) to be in at least 'good status' by 2027. 'Good status' represents the fourth level on a five-point scale from 'bad' to 'high'. Classification of surface waters is through assessment of the biological (fish, benthic invertebrates and aquatic flora), hydromorphological (e.g. bank structure) and chemical (e.g. oxygenation, phosphates and nitrates) status of water bodies. Groundwater is classified through the assessment of the quantitative (i.e. volume) and chemical status. At the time of submitting this thesis, the UK is currently in negotiations to withdraw from the European Union. It is understood that in the interim, all EU environmental laws will continue to be adopted by the UK Government (Environment Agency, pers comm). Despite the uncertainty over the future of this legislation, the Government has independently committed to a 25 Year Environment Plan (HM Government 2018). Within the plan, the Government recognises that 75% of all sediment loadings to aquatic environments originate from farming practices (Defra 2007, Bewes, Davey, and Keirle 2014) and aims to improve the ecological status of water bodies through enforcing regulations to reduce water pollution from agriculture. The provision of robust evidence to policy makers is crucial to ensure the preservation and continued improvement of freshwater environments whilst the long-term future of the environmental policy framework remains unclear. The results of this thesis will provide evidence to UK policy makers once the obligations under WFD cease and as new environmental legislations are implemented through UK Parliament.

Aquatic biomonitoring, the science of inferring the condition of the aquatic environment using the ecological community, has been standard practice for many years (Kolkowitz and Marsson 1908, Rosenberg and Resh 1993). Biomonitoring is conducted through the use of biotic indices, which allocate a score to each taxon based on their sensitivity to aquatic pollution. Aquatic macroinvertebrates are uniquely suited as biomonitors. They are widely abundant and ubiguitous in almost all environmental conditions, exhibit a large response diversity and are relatively easy to identify (Relyea, Minshall, and Danehy 2012). The most well-developed index in the UK, the Walley Hawkes Paisley Trigg index (WHPT) (Walley and Hawkes 1996), is used as a general indicator of aquatic health. This index is currently used by UK monitoring authorities to classify the biological status of water bodies. Biotic indices are dependent on the reliable allocation of sensitivity scores (Bonada et al. 2006). A variety of methods exist ranging from expert knowledge based to purely statistical approaches. It is important that any biotic index is thoroughly assessed before incorporation into national monitoring frameworks.

Physical methods of measuring both suspended and deposited fine sediment in rivers, while useful, can be time consuming, prone to errors and fail to integrate the conditions of the catchment, often only representing conditions at a single point in time. Furthermore, there is no globally agreed standard practice, and the multitude of methods available each measure a different component of the fine sediment system (e.g. superficial substrate or interstitial sediments and/or actively transported sediment). Given the realisation that fine sediment is a significant stressor of aquatic environments, and the problems associated with traditional physical methods of measurement, the last decade has seen the development of sediment-specific indices by scholars and management authorities. Among those developed for use in the UK are the Proportion of Sediment-sensitive Invertebrates (PSI; Extence et al. 2013), its empirical improvement (EPSI and EPSImixed; Turley et al. 2015, 2016) and the Combined Fine Sediment Index (CoFSI; Murphy et al. 2015). Disentangling the multifarious responses of aquatic biota to fine sediment is crucial to improving sediment-specific biomonitoring tools.

#### 1.2 Aims and Objectives

The title of this thesis is 'Quantifying the responses of macroinvertebrates to gradients of fine sediment pollution'. This overarching intention has been subdivided into three individual aims. Each aim is then achieved through a number of objectives and each objective aligns to a specific chapter within this thesis. The aims for this thesis are as follows:

## 1. Identify the main causal mechanisms involved in macroinvertebrate responses to fine sediment

- Objective 1.1 Review current literature (relevant to the research aims) to produce an overview of the knowledge on the fluvial sediment system, the responses of macroinvertebrates to fine sediment, and the importance of biomonitoring approaches in monitoring fine sediment (Chapter 2).
- Objective 1.2 Carry out a review using a systematic methodology to assess the weight of evidence for macroinvertebrate responses to fine sediment (Chapter 3).
- *Objective 1.3* Conduct a flume experiment to investigate the physical effects of fine sediment on macroinvertebrates (Chapter 4).
- 2. Compare and assess methods for quantifying suspended and deposited fine sediment in lowland gravel bed rivers
  - Objective 2.1 Carry out field work to compare different methods of measuring fine sediment (Chapter 5).
- 3. Test the response of macroinvertebrates to different metrics of fine sediment
  - Objective 3.1 Evaluate sediment-specific (e.g. PSI and CoFSI) and non-specific (e.g. WHPT) indices against different metrics of fine sediment (Chapter 5).

#### 1.3 Thesis structure

The structure of this thesis is outlined in Figure 1.1. The narrative literature review in Chapter 2 provides a detailed introduction to the sediment system, the

ecological impacts of fine sediment and monitoring practices for fine sediment including traditional physical methods and the move to biomonitoring style approaches. One of the roles of this chapter is to define the key concepts which are fundamental to the subsequent chapters. Chapter 3 follows a systematic approach to assess the evidence of macroinvertebrate responses to excessive fine sediment. Compared to Chapter 2, which provides a broad narrative overview, Chapter 3 addresses a more specific research aim by seeking to classify the types of macroinvertebrate responses to fine sediment. Chapter 3 assesses the wealth of published evidence to provide an 'evidence map'. Using a weight of evidence approach, the responses of macroinvertebrates to fine sediment are assessed based on the quality and rigour of the scientific study from which it originates. Higher quality studies receive a higher weighting and therefore contribute more to the overall evidence conclusion. Chapter 4 addresses key knowledge gaps identified in Chapter 2 and 3 by conducting a laboratory experiment to test whether fine sediment can cause physical damage, in the form of abrasion and clogging, on macroinvertebrate gill tissue. Macroinvertebrate cadavers are exposed to varying water velocities and concentrations of fine sediment in a recirculating flume. After exposure, a novel method of digital image analysis is used to determine the presence of physical damage from scanning electron microscopy images of individual gills. Chapter 5 reports results from a field study conducted to form an independent test of recently developed fine sediment-specific biomonitoring indices. Study sites are selected based on an extensive filtering process to minimise confounding factors. Within this chapter, different methods of measuring fine sediment are tested and compared. Some emerging methods in functional trait-based ecology and machine learning are explored in the novel analysis. Finally, Chapter 6 provides a summary of the key findings, draws conclusions from the collective results, and makes recommendations for future research.

The core of this thesis is embedded in ecological theory and applied practices. However, due to the cross-cutting nature of studying fine sediment in aquatic environments, this thesis will also span the disciplines of geomorphology and hydrology. This thesis incorporates the two broad epistemologies of

reductionism and holism (Figure 1.1). Whilst still sometimes ambiguous (Redfield 1988), reductionism attempts to isolate specific causes and effects, whereas holism takes a broader approach under which causal mechanisms are often uncertain (Trepl and Voigt 2011). The three results chapters (Chapter 3, 4 and 5) span both these approaches. Chapter 4, is particularly reductionist based on its controlled experimental approach designed to isolate two specific mechanisms hypothesised to control macroinvertebrate responses to fine sediment. The systematic review in Chapter 3 reviews evidence from both reductionist and holistic studies to provide an overall evidence synthesis. Finally, in Chapter 5, the field study takes a holistic approach. Whilst aiming to determine the overall effects of fine sediment, various site-specific abiotic and biotic factors will interact to determine the community at each site. The effects of these factors are disentangled with the help of a site-selection process and through statistical analysis.



Figure 1.1 – The relationships between the chapters and the different conceptual frameworks within this thesis. The increasing height of the central figure illustrates a move towards holism as opposed to reductionism.

# Chapter 2 – Fine sediment as a pressure of aquatic environments and progresses in monitoring approaches

#### **Chapter overview**

The erosion, transportation and deposition of fine sediment (<2 mm dia) are fundamental processes in the hydrogeomorphic cycle. However, increasingly intensive land management such as agriculture and construction, as well as inchannel sources such as channel incision, have elevated sediment levels beyond background levels. The delivery of excessive fine sediment to rivers can cause serious deleterious effects on aquatic ecosystems and is widely acknowledged to be one of the leading contributors to the degradation of rivers globally. This chapter begins by providing an overview of the fine sediment system including the sources and transport to river systems. Impacts of fine sediment are extensive and the effect on the ecological community can be complex. Fine sediment may be transported in suspension or deposited superficially or interstitially on or in the coarser river substrate. The multifarious impacts of sediment on macroinvertebrates are reviewed in this chapter.

Given the widespread impacts of fine sediment, measuring and monitoring its presence is required to evaluate the implementation of land management interventions and improve aquatic health. Physical methods of measuring fine sediment, while useful, can be time consuming, prone to errors and fail to integrate the conditions of the catchment, often only representing conditions at a single point in time (i.e. instantaneous rather than integrated over time) (Extence et al. 2013). Biomonitoring involves taking a community-wide approach to infer the environmental conditions at a given site. The fundamentals of biomonitoring lie within ecological theory, such as niche theory and disturbance response diversity. Aquatic macroinvertebrates are uniquely suited to their roles as biomonitors of stream health. Macroinvertebrate biomonitoring offers many benefits over traditional (physical) methods of

measuring fine sediment. Recognition of fine sediment as a significant pollutant of aquatic systems has led to the development of sediment-specific biomonitoring indices. This chapter ends by summarising the literature and highlighting the opportunities for developing the knowledge base on quantifying the response of macroinvertebrates to gradients of fine sediment pollution.

#### 2.1 Introduction

The aim of this chapter is to introduce the issues underpinning subsequent chapters and provide a synthesis of the relevant published literature to date. Key terms that will be used throughout the thesis will be defined in context. This chapter provides a narrative on the progress in the field, from the recognition of fine sediment as a significant stressor, to early studies quantifying individual or community effects, and lastly to the current development of sediment-specific biomonitoring and the emergence of species-trait-environment analyses.

#### 2.2 The sediment system

Erosion, transport and deposition of fine sediment are fundamental processes in the hydrogeomorphic cycle and river systems require a constant supply in order to function (Jones et al. 2012b). Diverse aquatic communities rely on the supply of fine sediment to provide suitable heterogeneous habitats and for delivery of particulate and dissolved organic matter (Collins et al. 2011). Fine sediments in river systems can be classified in two main fractions: deposited or suspended. The deposited fraction is the quantity of sediment that settles on the river bed. This deposited sediment can infiltrate into the substrate, a process known as colmation (Descloux, Datry, and Usseglio-Polatera 2014, Wharton, Mohajeri, and Righetti 2017). Depending on hydraulic conditions, sediment can transfer into the stream bed either vertically via the settling or turbulent diffusion of fine sediments from the water column, or horizontally through intragravel transport (Harper et al. 2017). The suspended fraction is the quantity of sediment that is held within the water column. The quantity of suspended sediment is intrinsically linked to the prevailing hydraulic conditions, catchment geology and geomorphological processes acting within a river system (Walling 2005).

The quantity of sediment transported downstream over a given period of time is described as the sediment load. Sediment can either be transported as bed load, suspended load or wash load (Table 2.1 and Figure 2.1). In terms of catchment management, the bedload is generally considered the most important fraction of sediment transport due to the effects on erosion and bedform change (Parker 1979, Talukdar, Kumar, and Dutta 2012). Despite comprising the smallest sediment particle sizes, the wash load can influence the optical properties of the water column (i.e. by increasing turbidity disproportionately for the same suspended sediment concentration as, for example, fine sand), reducing the depth to which light can penetrate (Waters 1995, Ziegler 2002). The total load, and the quantities contained in each component, will be influenced by the fluvial system (Ashworth and Ferguson 1989, Lane and Richards 1997) and can vary significantly, temporally and spatially. Mean standard suspended sediment concentrations (SSCs) in temperate rivers can vary three-fold inter-annually (Grove et al. 2015) with large seasonal variations as a result of high rainfall and flood events (Woodruff et al. 2001).

Sediment load	Description
compartment	
Bed load	Sediment particles saltating along the river bed.
Suspended load	The proportion of particles suspended in the water column.
Wash load	A component of the suspended load. It comprises the
	smallest sediment fractions (usually <2 $\mu$ m), including the
	colloidal fraction (particles 0.001-1 μm), which will typically
	always remain in suspension. However, colloids have a
	high surface charge and will readily form flocs, especially
	in the presence of organic matter, which can more readily
	deposit (Droppo et al. 1997).

Table 2.1 – A description of the three compartments of sediment load transportation; bed load, suspended load and wash load



Figure 2.1 – Fine sediment transport (black arrows) and deposition (grey arrows) processes in gravel beds. These processes occur in three distinct loads; the suspended, dissolved and interstitial load. Transport of fine sediment is affected by the change in velocity profile V(y) through the water and sediment column (right) (adapted from Casas-Mulet et al. 2017).

Largely a function of its source, the quality of sediment particles is closely associated with its impacts on the ecological community. The quantities of the organic and inorganic components of fine sediment can have important impacts on the biota in river systems (discussed in Section 2.3). Suspended sediment is estimated to be responsible for 27% of the global transfer of carbon to rivers (Meybeck 1982). Fine sediment also has the potential to interact with chemical elements and compounds, which can contribute to pollution of freshwater environments. The colloidal fraction of sediment is characterised by large surface areas and ionic charges which have the potential to attract and bind with other substances. Concentrations of heavy metals have been found to be 100-10,000 times higher in the sediment than in the water column (Yi et al. 2008). Fine sediments have also been shown to sorb pesticides (Gilliom and Clifton 1990, Gao et al. 1998), nutrients such as nitrates and phosphates (Tournoud et al. 2005) and polychlorinated biphenyls (PCBs) (Walling et al. 2003). The term Persistent Organic Pollutants (POPs) is used to describe chemicals that sorb strongly to solids due to their hydrophobic and lipophilic nature (Jones and de Voogt 1999). Fine sediments in aquatic systems are a
significant sink for POPs and represent an important pathway into the food web as the contaminants become bioavailable to aquatic organisms (Rainbow 1995). The presence of sediment-associated contaminants is a significant problem closely associated with fine sediment delivery to aquatic systems. However, consideration of the wealth of evidence related to ecotoxicological effects is beyond the scope of this thesis.

Excessive fine sediment delivered to aquatic environments is a significant threat to ecosystem health (Yule, Boyero, and Marchant 2010). Defining 'excessive' fine sediment is complex. At the most basic level, and the definition implied in this thesis, 'excessive' can be defined as the quantity over and above that of natural background levels, i.e. the supply and delivery of sediment has been altered or enhanced in some way, usually anthropogenically (Bilotta et al. 2012). Nonetheless, quantifying 'background levels' of fine sediment is exceptionally difficult. From a paleolimnological perspective, Foster et al. (2011) defines excessive quantities as sediment yields that are significantly greater than pre ~1940 levels (prior to the most dramatic increase in sediment yields which occurred after 1945 according to Foster et al. 2006). Background fine sediment levels will also naturally vary depending on spatial variation and key catchment drivers (e.g. geology and catchment land use). Therefore, delineating the empirical quantities of fine sediment as a result of natural variation to that of enhancement from anthropogenic activities is still poorly understood by both academics and river managers.

Fine sediment is often described as a diffuse pollutant in aquatic environments and the term 'fine sediment pollution' is used throughout this thesis to describe the excessive delivery and retention of 'fines' (fine sediment). Increasingly intensive agricultural land management, construction, mining, deforestation, and in-channel modifications, leading to bank erosion and channel incision, are some of the main anthropogenic sources leading to increased sediment loads of rivers (Owens et al. 2005, Collins et al. 2009b, Yule, Boyero, and Marchant 2010). Since industrialisation, rapid population increase and development of specialised agricultural machinery has led to more intensive land management practices (Zhang et al. 2014). Before 2005, the European Union Common

Agricultural Policy subsidies were coupled with production, therefore the incentive to intensify was driven not just by external profits but from an increased subsidy payment. More recently, in many parts of the United States, large areas are being converted to agricultural land for the cultivation of biofuels (Klco 2008). Soil carried off in rainwater or by irrigation from intensively farmed agricultural land will inevitably end up in aquatic systems.

Excessive fine sediment delivery, when coupled with relatively low transport capacity of lowland rivers (Naden et al. 2016), results in channels choked with fine sediment causing significant impacts on aquatic communities. As a result of this, fine sediment is considered to be a significant pollutant to aquatic systems globally (Owens et al. 2005). However, the impacts of soil erosion from land sources extend beyond ecological impacts to aquatic communities. Soil degradation in England and Wales has a total economic cost of an estimated £1.2 billion per year (Graves et al. 2015). 'On-site' costs to farmers and landowners include yield losses or costs incurred through mitigating soil erosion. Costs incurred by wider society are those which occur 'off-site' such as flooding of properties as a result of rapid run-off from cultivated hill-slopes or effects on drinking water quality. Increased sediment delivery to river systems can cause significant implications for river regulation. The results are serious: flooding, navigation blockages, and large build ups at weirs and dams leaving channels requiring regular maintenance, such as dredging or dam flushing which can deliver large slugs of sediment downstream (Owens et al. 2005). Effective monitoring practices can more efficiently identify areas affected by fine sediment before it becomes a significant problem (i.e. before the aquatic community has become degraded). This in turn can help river regulators (e.g. the Environment Agency) advise land managers to implement mitigation measures to reduce sediment input to rivers. Thereby, benefitting both river environments and the wider community.

### 2.3 Ecological impacts of fine sediment

The ecological impact of fine sediment will be a function of its source, quantity, timing of delivery and retention (Murphy et al. 2015). Effects of fine sediment on

fish are well documented because of their commercial and economic importance (Wood and Armitage 1997). However, the huge functional diversity of macroinvertebrates makes their response to environmental stressors complex and despite being less economically significant than many fish species, they are important components of aquatic ecosystems. Macroinvertebrates are important engineers of aquatic environments and can regulate processes from both top-down and bottom-up controls. Invertebrate grazers have been shown to control both the biomass and taxonomic composition of their algal food source (Lamberti and Resh 1983, Hillebrand et al. 2002, Cibils-Martina et al. 2019). In addition, macroinvertebrates can have significant impacts on abiotic conditions in river environments such as the storage and transport of fine sediments (see 'biota' box in Figure 2.2) (Albertson and Allen 2015, Wilkes et al. 2019). Macroinvertebrate behaviours, such as feeding activities (Pringle et al. 1993, Nunokawa et al. 2008) or burrowing (Mermillod-Blondin et al. 2003, 2004, Holdich et al. 2014), can exert controls on fine sediment. Macroinvertebrates also provide a significant food source for riverine fish species (Vidotto-Magnoni and Carvalho 2009), supporting their economic value. Aquatic invertebrates with terrestrial adult life stages provide important subsidies to the riparian zone as a food source for terrestrial organisms (Paetzold, Schubert, and Tockner 2005). Lastly, aquatic macroinvertebrates are important biomonitors of ecosystem health which is critical to the focus of this thesis. Their importance and suitability as biomonitors will be covered in Section 2.4.2.

Aquatic organisms rely on the supply of fine sediment to provide suitable habitats and for delivery of particulate and dissolved organic matter (Collins et al. 2011). Macroinvertebrates have a wide-ranging association with the river bed including burrowing, hiding or attachment which instils a requirement for habitat heterogeneity to meet the demands of local populations (Tachet et al. 2010). However, excessive sediment delivery can have serious deleterious effects on aquatic biota. Macroinvertebrate responses to fine sediment represent a complex mix of direct and indirect effects (Jones et al. 2012b). There is a range of literature citing that both deposited and suspended sediment

can result in changes to the biotic community. Different components of the macroinvertebrate assemblage will respond to different aspects of sediment pollution depending on their relationship with the substrate, feeding behaviours and other functional traits (Culp, Wrona, and Davies 1986, Angradi 1999, Suren and Jowett 2001, Larsen and Ormerod 2010). Most components will respond negatively but some will respond positively (e.g. Oligochaeta; Cover et al. 2008, Wagenhoff, Townsend, and Matthaei 2012, Davis et al. 2015) and therefore the relationship is not as simple as an inverse association between sediment quantity and abundance or richness of taxa. Instead, there is a complex web of interactions and effects (Figure 2.3). The next Sections (2.3.1 and 2.3.2) summarise both historical and emerging evidence of macroinvertebrate responses to fine sediment and the mechanisms through which this can drive change to the community.

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Figure 2.2 – Potential pathways from source to impact of fine sediment in river networks (from Wilkes et al. 2019). Factors affecting sediment yield, delivery pathways, transport, and storage ultimately influence the potential ecological responses which in turn can create feedback loops and exert upward controls on sedimentological processes.



Figure 2.3 – A conceptual model of the mechanisms through which fine sediment can affect macroinvertebrate community composition. The model has been broken down into the two main fractions of fine sediment; suspended and deposited (adapted from Jones et al. 2012b).

## 2.3.1 Suspended sediment

Increased SSC can create highly turbid water columns. Turbidity can be defined as 'a decrease in the transparency of a solution due to the presence of suspended and some dissolved substances, which causes light to be scattered, reflected, and attenuated rather than transmitted in straight lines; the higher the intensity of the scattered or attenuated light, the higher the value of turbidity' (Ziegler 2002, p1). A higher concentration of suspended sediments within a river system results in increased light attenuation and decreased depth to which light can penetrate (Waters 1995). In turbid waters, the compensation depth is reduced (the level at which photosynthesis equals respiration in plants) (Batiuk et al. 1992) which constrains photosynthesis to the upper levels of the water column (Berry et al. 2003). Photosynthesising organisms are the key component of primary production and reducing their activity has a cascading effect on upper trophic levels. Various studies have shown links between suspended solids, turbidity and primary production (Nieuwenhuyse and LaPerriere 1986, Klco 2008, Jones et al. 2012b). Reduced rates of photosynthesis or chlorophyll *a* concentration (often used as a proxy for photosynthetic activity) as a result of increased suspended solids have also been shown in Lloyd (1987), Rivier and Seguier (1985), and Suren and Jowett (2001). Primary production is the foundation of trophic webs, and any reduction of this process will reduce the flow of energy to higher trophic levels (Izagirre et al. 2009, Aspray et al. 2017).

An increase in turbidity can also affect behaviour and activity of organisms that use visual searching behaviours. There is an observed effect on fish that rely on visual search strategies during increased turbidity such as the reduction in prey consumption in striped bass (*Morone saxatalis*; Breitburg 1988) and a change in prey selectivity to slower moving species coupled with a reduction in feeding rate in largemouth bass (*Micropterus salmoides*; Shoup and Wahl 2009). However, some fish may benefit from the reduced visual acuity of their prey species during turbid conditions therefore increasing foraging ability (Gregory and Northcote 1993). Most research analysing searching behaviour and effects on visual searching have been carried out on fish species. However, it is possible that predatory invertebrates that also rely on visual searching behaviours could be affected, such as adult and larval diving beetles (Coleoptera: Dytiscidae), adult bugs (Heteroptera: Nepomorpha), and larval dragonfly and damselfly (Odonata) (Klecka and Boukal 2012).

There is limited evidence of direct physical effects of fine sediment, such as clogging and abrasion, on macroinvertebrates. Suspended sediments, particularly clays and the colloidal fraction, can build-up on organs, disrupting the normal functioning of gills, osmoregulation and feeding apparatus (Jones et al. 2012b). Experimental evidence has shown organisms with exposed feeding apparatus spend an increased portion of time and energy cleaning their feeding apparatus or expelling unwanted ingested particles (pseudofaeces) when suspended solids concentrations are high (Arruda, Marzolf, and Faulk 1983,

MacIsaac and Rocha 1995, Iglesias et al. 1996). The frequency of these behaviours increases with increased particle size and sediment load (Runde and Hellenthal 2000). Although limited evidence exists, it is theorised that aquatic organisms can be impacted from the abrasive effects of particles, either saltating or carried in suspension, which could cause dislodgement or damage to their body parts (Culp, Wrona, and Davies 1986). Exposed non-chitinous tissue (e.g. gills or feeding apparatus) could be damaged by fast flowing sediment, particularly the larger fraction that becomes mobilised in heavy storm flows (Jones et al. 2012b).

The abrasion theory is in part explained by behavioural responses observed under high sediment concentrations e.g. retraction of feeding apparatus (Kurtak 1978), inhibition of feeding from rapid gut filling (Gaugler and Molloy 1980) and a switch in feeding modes (e.g. from filtering to grazing; Voelz and Ward 1992). Evidence such as this is used by some researchers to demonstrate the effects of abrasion (e.g. Jones et al. 2012b). However, the evidence is potentially spurious as such behavioural changes could be explained by other mechanisms. For example, switching feeding modes in high suspended concentrations could be because, as the number of suspended sediment particles increases, the relative concentration of particulate organic matter decreases and therefore filter feeding is not effective and switching to grazing on periphyton is more efficient. This is still an effect of increased sediment in suspension, but not a direct result of abrasive forces acting on organisms.

Another consequence of suspended sediment, sometimes attributed to abrasive forces acting upon stream benthos, is macroinvertebrate drift. Drifting is a natural dispersal process in aquatic systems and varies spatially and temporally and with diel patterns (Svendsen, Quinn, and Kolbe 2004). Despite the exact mechanisms behind diurnal drift remaining unclear, drifting is a common response to disturbance (Mackay 1992). This sublethal effect has been shown to affect density, diversity and community structure of invertebrates and effects of this kind have the potential to cascade throughout the trophic web. Fine sediment addition can be more influential in eliciting drift responses than other forms of pollution (e.g. glyphosate herbicide; Magbanua et al. 2016). The term

'catastrophic drift' is used to describe the marked increase in drift (i.e. over and above that of natural drift patterns) in response to disturbances such as floods or pollution events over and above that of natural drift patterns (Lauridsen and Friberg 2005, Gibbins, Vericat, and Batalla 2007). Such 'catastrophic drift' of macroinvertebrates has been demonstrated from experimental additions of fine sediment (both suspended and deposited) resulting in reductions of benthic macroinvertebrate density of 30-60% (Culp, Wrona, and Davies 1986, Suren and Jowett 2001, Larsen and Ormerod 2010). However, drift response has been shown to be species-specific and will differ depending on an organism's relationship to the substrate (Runde and Hellenthal 2000, Suren and Jowett 2001).

### 2.3.2 Deposited sediment

Studying the effects of deposited sediment is complex because of the potential influence of compounds associated with sediment such as nutrients, metals, organic matter, POPs, as well as, the shape, size and volume of sediment deposited. Together with the longer residence time of sediments deposited on the river bed compared to the transience of suspended sediments, this results in a multitude of response mechanisms. Maintaining flow in aquatic environments is essential for supplying fresh nutrients, replenishing gases and removing waste. The settling and infiltration of fine sediment by colmation clogs the spaces between gravels reducing interstitial water flow critical for the exchange of gas in these pore spaces (Figure 2.4), thereby restricting the supply of oxygen to benthic organisms and the removal of excreta (Owens et al. 2005). Numerous studies detail the effect of sediment deposition on the incubation and survival of fish eggs, particularly salmonids because of their economic significance (Bruton 1985, Greig, Sear, and Carling 2005, Jensen et al. 2009, Sear et al. 2017). Sediment deposition can affect fish directly by reducing spawning habitat, smothering eggs, reducing overwintering and blocking fry emergence, and indirectly by altering invertebrate species composition, i.e. prey abundance (Sear 1993, Kemp et al. 2011, Relyea, Minshall, and Danehy 2012). Fine sediment deposition can reduce primary production by smothering the benthos and directly limiting light penetration to

primary producers (Vermaat and Bruyne 1993, Aspray et al. 2017). Indirect effects altering fish population and primary production can result in cascading effects on macroinvertebrate communities.



Figure 2.4 – An image of a section of the River Colne (Essex) with a high quantity of deposited sediment which has infilled and smothered the underlying gravel bed (a) and River Misbourne (Buckinghamshire) channel with clean visible gravel bed and low overlying fine sediment (b). Both rivers are typical lowland rivers. Flow direction left to right in both images.

Sediment deposition can directly affect macroinvertebrates through burial. The extent of this effect will depend on the species, sediment size and burial depth (Dobson, Poynter, and Cariss 2000, Wood, Vann, and Wanless 2001, Wood et al. 2005). The ability of individuals to excavate themselves from sediment burial can provide an indication of their sensitivity to fine sediment. Most recently, Conroy et al. (2018) ranked factors affecting species responses to burial as: burial depth > sediment size class > species source (i.e. upland or lowland). No effect of body size on species response could be detected. This is in contrast to previous evidence which established body size as an important factor in determining sensitivity to fine sediment (Gayraud and Philippe 2001,

Wagenhoff, Townsend, and Matthaei 2012, Descloux, Datry, and Usseglio-Polatera 2014). It is important to recognise that each respective burial experiment utilised different test species and experimental conditions, potentially inhibiting the use of generalisations on the effects of sediment by burial. Full descriptions of each study can be found in Table 2.2.

While organic matter in natural systems is vital as a food source to benthic organisms, disturbance to this critical input can alter the trophic system. An increase in organic matter can increase metabolic rates at the ecosystem level, particularly through bacteria decomposing the organic material, which increases the requirement for oxygen (biological oxygen demand) (Bjornn and Reiser 1991). The response of macroinvertebrates to reduced oxygen environments has been well studied. Most research around increased organic matter and subsequent decreased oxygen in aquatic systems has mostly been focussed on sewage effluent. Murphy et al. (2015) indicate that taxa are unlikely to be able to distinguish between the various sources of organic matter that cause reduced oxygen stress. Thus, at least some of the impacts of fine sediment could be similar to those of organic pollution. Flocculation of organic matter facilitates the settling and storage of particles on the stream bed (Burban et al. 1990). These deposited particles can cause 'capping or blocking' of intra-gravel flow which exacerbates the effect of smothering from inorganic particles and reduced oxygen from organic particles (Owens et al. 2005). Therefore, organisms with a tolerance for low oxygen environments, such as the families Asellidae, Vivparidae and Sialidae (Surber and Bessy 1974, Jones et al. 2009), may tend to dominate in areas affected by sediment deposition (Hinchey et al. 2006). Furthermore, organic matter content was found to be the primary gradient of sediment pollution effecting invertebrate community structure in a large-scale field study (Murphy et al. 2015).

Table 2.2 – Summary of burial experiments from existing literature. Upland/lowland distinction is based on the boundaries described in Conroy et al. (2018). This item has been removed due to third party copyright. The unabridged version of the thesis can be viewed at the Lanchester library, Coventry University

Gravel river beds can act as a short-term storage for sediments on the bed surface (Rosenberry and Healy 2012) or provide long-term storage within the gravel matrix (Thoms 1994, Heppell et al. 2009). The transport of sediment associated contaminants and POPs into river systems was described in Section 2.2. The presence of these substances can persist in the gravel bed matrix for long periods or become bioavailable to stream organisms (Eljarrat et al. 2004). Metabolism of POPs is slow and their transfer through food chains has been well studied (e.g. Yi et al. 2008). The effects of sediment associated contaminants crosses a disciplinary boundary in to the field of ecotoxicology. Despite the importance of these contaminants when considering the impacts of fine sediment in river systems, their effects are beyond the scope of this thesis.

Considering rivers as dynamic systems, single stressors rarely occur in isolation. Acknowledging the direct link between agriculture and fine sediment inputs to river systems (see Section 2.2), fine sediment stress can regularly be coupled with substances derived from fertilisers or herbicides and pesticides. Several studies have examined the impacts of fine sediment in a multi-stressor environment using mesocosms and full factorial experimental designs. Studies by Davis et al. (2018, 2019) showed that the effects of nitrogen and phosphorous were relatively weak compared to fine sediment addition and that the community could not recover while sediment was still present at elevated levels. Magbanua et al. (2016) showed fine sediment to have greater impacts on eliciting macroinvertebrate drift and adult emergence than a glyphosate-based herbicide. The implications of these studies point towards fine sediment as the 'master stressor' of macroinvertebrates in river systems and priority should be given to managing, and understanding the effects of, sediment inputs (Davis et al. 2019).

### 2.4 Monitoring fine sediment

The environmental impacts of fine sediment are pervasive. It is important that environmental managers employ effective monitoring practices to efficiently identify areas effected by fine sediment. This section will outline methods of

monitoring fine sediment including traditional physical methods which aim to directly quantify the mass or concentration of fine sediment, the benefits of biomonitoring approaches and the global development of fine sediment-specific monitoring approaches. This section will partly illustrate the historic use of physical methods and basic biomonitoring approaches up to the current academic literature using biotic indices and emerging trait-based approaches.

## 2.4.1 Physical methods

Traditionally, a multitude of physical methods have been employed to quantify suspended or deposited fine sediment in river systems. These methods span a large gradient of cost, time, effort and complexity. Furthermore, different techniques will measure slightly different components of fine sediment (e.g. deposition rate, organic content, turbidity etc.) which makes comparisons between methods challenging. This section will discuss some of the most common physical methods of measuring suspended and deposited sediment.

## 2.4.1.1 Suspended sediment

Suspended sediment is typically measured as a concentration per volume of water (e.g. mg l<sup>-1</sup>). A known volume of water is sampled from a river, filtered, dried and the contents weighed to approximate an SSC (UK Standing Committee of Analysts 1980, Gray et al. 2000). This process is time consuming, can be expensive if a large number of samples are required and necessitates off-site sample processing using laboratory facilities (Bilotta and Brazier 2008). The light scattering properties of water, measured using turbidity, is often used as a surrogate for SSC (i.e. the higher the turbidity value, the higher the SSC). Turbidity can be easily and cheaply measured in lentic systems using a Secchi disk. The Secchi depth is the depth, when lowered into the water column, at which the disk is no longer visible. The light attenuation coefficient of Photosynthetically Active Radiation (PAR), an ecologically relevant metric, can then be extracted from the Secchi disk depth value (Padial and Thomaz 2008). This is a quick and low-cost method but will also have high operator variability and disturbances to the water surface when operating the Secchi disk make it unsuitable for lotic systems (Larson and Buktenica 1998). Suspended sediment,

and hence turbidity, are characterised by high levels of variability linked to hydraulic conditions. These instantaneous methods only measure SSC or turbidity at a single time point, thus failing to capture variations in SSC over time.

Time integrated turbidity loggers are an improvement on the issues associated with taking physical samples to quantify SSC directly. Turbidity loggers use properties of optical light scattering to determine turbidity measurements expressed as Nephelometric Turbidity Units (NTU) (Lewis 1996). However, they do not directly represent SSCs. The readings can be skewed by scattering of other particles including algae, plankton, organic matter, microbes, air bubbles and other fine insoluble particles and flocculated particles. This can lead to underestimates of absolute SSCs unless a site-specific calibration can be obtained (Lawler et al. 2006). Furthermore, variation in sensor type can result in up to five-fold differences in measured turbidity levels (Rymszewicz et al. 2017). Acoustic Doppler Meters measuring backscatter can also be used to measure SSC integrated over time and space which can provide more information than turbidity meters or probes, but still require complex calibrations and will also be affected by over/underestimates of readings.

Despite the inaccuracies of suspended sediment measurements, some international guidelines have been developed setting SSC targets as the required standard. A thorough search of the literature yielded only three international directives which have incorporated this measure into environmental policies (Table 2.3). This limited application could be reflective of the inaccuracies of applying blanket guidelines of SSC which is prone to fluxes heavily dependent on flow dynamics. The Canadian Environmental Quality Guidelines (CEQC) attempts to assuage this by defining separate guidelines at high and low flows. The European Union Freshwater Fish Directive (78/659/EEC) (2006/44/EC) which previously stated a guideline standard of ≤25 mg l<sup>-1</sup> annual average concentration (except in exceptional circumstances such as storms or droughts) was repealed in 2009 when it was replaced by the European Water Framework Directive (WFD) (2000/60/EC) (2008/105/EC). The WFD does not contain SSC standards. The Australian and New Zealand

Guidelines for Fresh and Marine Water Quality (ANZECC 2000) set turbidity standards for different water body types in Australia (individually by State) and New Zealand. Arguably more unreliable than SSC, the uncertainties of using turbidity as a proxy have already been covered in this section. The application of meaningful sediment targets continues to draw scientific debate (Collins et al. 2011) and recommendations for the development of any future guidelines focus on the implementation of a holistic approach such as the inclusion of catchment drivers, sediment regimes and channel morphology, coupled with ecologically relevant responses (Bilotta and Brazier 2008, Collins et al. 2011).

Directive/Regulation	Region/ Country	Standard
Freshwater Fish Directive (78/659/EEC) & (2004/44/EC) [DIRECTIVE HAS BEEN REPEALED]	European Union	≤25 mg l <sup>-1</sup> annual average concentration apart from exceptional conditions (e.g. floods and droughts)
Canadian Environmental Quality Guidelines (CEQC) for Protection of Freshwater Aquatic Life (CCME 1999)	Canada	At low flow (above background): • <25 mg I <sup>-1</sup> (<24 hrs exposure) • <5 mg I <sup>-1</sup> (1-30 days exposure) At high flow (above background): • <25 mg I <sup>-1</sup> (when background 25- 250 mg I <sup>-1</sup> ) • <10% of background concentration (when background >250 mg I <sup>-1</sup> )
US EPA (2007); US Clean Water Act (1972)	USA	Suspended and settleable solids should not reduce the depth of the compensation point (see Section 2.3.1 for definition) for the photosynthetic activity by >10% from the seasonally established norm for aquatic life. Total maximum daily loads (TMDLs) to be defined on a state-by-state basis

Table 2.3 – International guidelines for SSC (based on Table 4 from Bilotta and Brazier 2008).

#### 2.4.1.2 Deposited sediment

Deposited sediment is normally measured as a volume or mass of sediment per unit area (or per unit volume for infiltration), depending on the method used, can be quantified over a unit of time (i.e. deposition rate). Taking grab samples or sediment cores from river beds can be a relatively simple and basic method of obtaining a fine sediment mass per unit area. However, both of these methods present problems with disturbance during mechanical removal which can lead to loss of the finest fractions during extraction (Thoms 1992), and are often only suitable for exposed drained channel bars (Carling and Reader 1981). The coring method has been improved by freezing the bed in situ by injecting liquid nitrogen or CO<sub>2</sub> thus freezing the adjacent hyporheic water and gravel matrix (e.g. Descloux et al. 2010). Freeze-coring has been shown to be a more accurate technique as grab-sampling can underestimate the fine sediment proportion by mass (Thoms 1992, Milan et al. 1999). However, bed fabrics can become disrupted when the coring probe is driven into the sediment and it is also a relatively destructive method not suitable for extensive or frequent surveys (Kondolf, Lisle, and Wolman 2003).

Measuring both surface and infiltrated sediment instantaneously can be done via the disturbance method. This method, also called the resuspension method, was first described by Lambert and Walling (1988) and later developed by Collins and Walling (2007a, 2007b) then Duerdoth et al. (2015). The method uses an open-ended hollow cylinder of known diameter pushed within the gravel bed to achieve an adequate seal from the surrounding flow. Once a seal is achieved, the overlying water is vigorously agitated manually without touching the river bed in order to bring unconsolidated surface sediment into suspension and the overlaying water is sampled to determine the concentration and mass released (i.e. total surface sediment). The process is then repeated including agitation of the top 100 mm of the gravel bed to raise interstitial fine sediment into suspension thus measuring both the surface drape and the infiltrated (subsurface) sediments combined (i.e. the total sediment). The water samples taken from both the surface and subsurface agitation can then be recovered from suspension in the laboratory allowing for further analysis (e.g. particle size,

sediment-associated nutrients and contaminants). Recently assessed for its accuracy, this method showed low variance associated with operator or other within-site differences (Duerdoth et al. 2015, Conroy et al. 2016b).

Coring, freeze-sampling or the disturbance methodologies do not allow quantification of the rate of deposition. This can be achieved using sediment traps which lie in situ in the gravel bed. When the traps are installed, they are filled with clean gravel (gravel larger than 2 mm) so after removal, the trap contents can be sieved and the quantity of particles <2 mm represent the accumulation rate of fine sediment over the installation period. There are numerous methods of sediment trap design which usually vary in their ability of sediment to ingress vertically or horizontally into the trap. Several studies have demonstrated that horizontal (lateral) sediment transport can account for a considerable proportion of total sediment transport (e.g. Carling 1984; Sear 1993; Mathers and Wood 2016). Harper et al. (2017) discusses the complexities of measuring fine sediment using bed trap methods, how the use of clean gravel is contrived and suggests that the results must always be interpreted with care.

Quantifying the surface drape (the overlying sediment in the upper layer of the gravel bed) of fine sediment requires an assessment of the entire reach due to natural variation in sediment storage across mesohabitats (Sear 1996). Visual estimates, described in the River Habitat Survey Field Survey Guidance Manual (Environment Agency 2003) involve the operator estimating the percentage substratum composition over a given reach of the river. Substrates are recorded using seven size categories (Table 2.4). The percentages of sand, silt and clay are then combined to provide an estimate of fine sediments. However, visual estimates can be subjective with up to 40% of between user variability (Duerdoth et al. 2015). It must also be considered that this method, which only allows quantification of the surface drape and not the extent of sediment retention within the interstitial spaces, may not be the most accurate method when quantifying fine sediment (Duerdoth et al. 2015). However, several studies have supported the accuracies of the visual estimate method. Both Zweig and Rabeni (2001) and Glendell et al. (2014) found that the measure of

embeddedness and visual estimates were highly correlated with one another, implying that visual estimates are consistent with embeddedness below the surface drape. Conversely, Bunte and Abt (2001) suggest that visual fines could be an underestimate of subsurface sediment due to vertical stratification of sediments resulting in finer sediments in the subsurface than the surface. Several studies have found that the total percentage of fines from visual estimates explained the most variation in macroinvertebrate assemblage (Sutherland, Culp, and Benoy 2012, Glendell et al. 2014, Conroy et al. 2016a).

Table 2.4 – Sediment size categories and simple field descriptions for visual estimates of fine sediment (adapted from Environment Agency 2003; Shuker et al. 2017).

Category	Size	Field description
Bedrock		Exposed (solid) bedrock
Boulder	>256 mm	Larger than head size
Cobble	64 – 256 mm	Half-fist to head size
Gravel-	2 – 64 mm	Particles clearly visible to the naked eye from
Pebble		several metres
Sand	0.0625 – 2 mm	Loose and crumbly material, visible to the
		naked eye from 1 m
Silt	0.00195 –	Loose, crumble material but individual
	0.0625 mm	particles difficult to see with the naked eye
Clay	<0.00195 mm	Sticky, cohesive material

As visual estimates only accurately quantify surface drape (compared to subsurface ingress of fine sediments) this could provide an indication of the fraction of fine sediment pollution that is most likely to affect the macroinvertebrate community. Several efforts have been made to improve the accuracy of visual assessment methods. Clapcott et al. (2011) developed a protocol using a bathyscope (underwater viewer) to reduce subjectivity when taking in-stream visual estimates. This involves the operator estimating the %

fine sediment within the gridded area of the streambed observed through the bathsyscope lens. The protocol recommends the process is carried out at four random locations across five random transects. Clapcott et al. (2011) recognises that this method is difficult to use in fast, shallow flows as the bathyscope can cause turbulence, entraining fine sediment. Turley et al. (2017) developed a method using digital image analysis of photographs taken of the river bed (at a known depth) to calculate the percentage of sediment coverage. This method helps to reduce operator bias when making estimates of surface sediment cover.

In summary, whilst physical methods of measuring fine sediment coverage of the river bed or storage within the substrate are important, they can be time consuming, destructive, prone to errors and often only representative of a single point in time. Furthermore, these methods do not take into account the ecological impacts of fine sediment (Turley et al. 2015, Murphy et al. 2015). The next section will discuss the use of macroinvertebrates as biomonitors of stream health and the benefits of these methods over traditional, direct physical measurements.

### 2.4.2 Macroinvertebrates as biomonitors

The theory of evolution by natural selection (Darwin 1859) is critically linked to the interaction between the characteristics of the individual and its environment (Begon, Townsend, and Harper 2006). Adaptation is inherently connected with the concept of ecological fitness which is a '*measure of competitive success, the tendency of an organism to increase the representation of its genes in successive generations*' (Peacock 2011, p100). Fundamental drivers at the genetic level have shaped and moulded organismal communities with diverse physical, phenological, biological and ecological diversities. Although driven by the environment, communities of organisms will also contain species which are distinct or specialised as no habitat is entirely homogenous (Begon, Townsend, and Harper 2006). Niche differences between species (interspecific), as opposed to within species (intraspecific), results in a gradient of responses within a community to disturbance events, i.e. 'response diversity' (Elmqvist et

al. 2003). This has provided scientists with the ability to predict ecosystem health using local biotic assemblage as opposed to taking direct measurements of the abiotic environment, such as pH, nutrients or chemicals (e.g. herbicides). This section applies the fundamental principles of ecological theory to describe how evolution and adaptation underpins the use of organisms as a proxy to monitor environmental quality and why aquatic macroinvertebrates are uniquely suited to this role (Johnson, Wiederholm, and Rosenberg 1993).

Understanding specialism and diversity is important in quantifying organismal response. At the most basic level, this can begin with the physical environment in which an organism lives, i.e. the habitat. Although commonly used interchangeably with 'habitat', Grinnell (1917) was the first to use the term 'niche' which was later defined by Elton (1933) as 'how' and not 'where' an organism lived. This was further developed and today the most widely used definition of a niche is given by Hutchinson (1957) as the total range of environmental conditions under which a species can exist. Referred to as the Hutchinsonian niche, this 'space' can be defined as an n-dimensional hypervolume (Stevenson 1982). The Grinellian definition of 'niche' refers to an organism's tolerances and preferences rather than the actual physical environment in which it lives (Whittaker, Levin, and Root 1973). However, community ecology is considerably more complex than this definition and multifarious interactions within communities, such as predation, competition and facilitation, can constrain organismal succession. Hutchinson (1957) expanded this definition further; an organism has a larger ecological niche in the absence of competitors and predators. Thus, Hutchinson's concept recognises the fundamental niche (the overall potentialities of a species) and the realised niche (the more limited spectrum of conditions and resources that allow it to persist, even in the presence of competitors and predators). It is important to distinguish between habitat and niche when describing interactions within communities because no two species in a stable community with limited resources can share the same niche (Whittaker, Levin, and Root 1973, Wiley 1978). There is evidence that in lotic systems, abiotic factors are more important than biotic factors in regulating community structure (Ledger and Hildrew 2000). Therefore,

interactions in ecological communities can rarely be understood until the community is studied as a whole rather than at individual species level.

There is no single method that can be used to quantify the effects of pollution on an organism. A suite of techniques is used depending on the stressor such that it has stimulated an entire subfield of work known as ecotoxicology. The most basic method would be quantifying mortality in the field after a known pollution event has occurred (e.g. mass salmon deaths from acid pulses after spring snow melts; Hesthagen 1989). Often for regulatory purposes, laboratory experiments seek to quantify the lethal concentration ( $LC_{50}$ ) or effective concentration ( $EC_{50}$ ) that will kill half the sample population during a given amount of time (Trevan 1927). This method has been used historically to determine the toxicity of drugs, pesticides and other chemicals. However, not all pollution events result in immediate mortality, at least not of the entire ecosystem, but still have significant effects on the health of the ecological community. Early detection of pollution is critical for management practices and methods of detecting non-lethal levels must be employed by integrating the response of the aquatic community e.g. biomonitoring.

Freshwater biomonitoring is the 'science of inferring the ecological condition of rivers, lakes, streams and wetlands by examining the organisms that live there' (Claro, Oliveira, and Rico-Gray 2009, p63). Instead of measuring environmental variables directly, organisms are used as a proxy. Biomonitoring is generally used as a catch all term but can be broken down into two different applications; bioindicators and biotic indices. The term bioindicator is applied to individual organisms that are used to quantify the pollutant levels across various spatial or temporal gradients. Physiological changes (sublethal response) in response to a contaminant can be used for quantification. For example, the presence of imposex in *Nucella lapillus* can be used as a bioindication of tributyltin pollution (Bigatti et al. 2009) or analysis of tissue samples to provide an indication of bioavailability of heavy metals in an ambient habitat (e.g. heavy metals; Rainbow 1995).

A community wide approach of biomonitoring involves the use of biotic indices. An index system works by assigning each taxon a score based on their sensitivity/tolerance to a particular pressure. The sensitivity scores can either be derived from expert knowledge, through a multivariate approach, or a combination of both (Birk et al. 2012). The habitat is manually surveyed and the scores of each taxon present will relate to a scale of ecosystem health. The principle behind this method is that different organisms will have different tolerances to pollutants. The main advantage of this type of monitoring is that the community reflects transient events that could be missed by direct environmental monitoring to provide an integrated view of the ecosystem as a whole (Rosenberg and Resh 1993, Bonada et al. 2006, WFD-UKTAG 2014).

Many biotic indices have been developed for use in aquatic environments. Aquatic macroinvertebrates are one of the most diverse taxonomic groups on earth, particularly because they are almost ubiquitous in freshwater environments (Rosenberg and Resh 1993). Macroinvertebrates also integrate the conditions over time during their life cycle. Lower trophic levels such as algae and phytoplankton have a rapid turnover time that may not reflect longer temporal scales of environmental health conditions and rapid identification is not possible (Resh 2008). Higher trophic level organisms, such as fish, are longerlived and their response time to non-point source pollution may be too long for monitoring purposes. Furthermore, fish and other vertebrates are highly mobile, or even migratory, so may not be representative of the study area (Relyea, Minshall, and Danehy 2012). In addition to this, macroinvertebrate's diversity, abundance and prevalence in aquatic systems worldwide and relative easiness to identify (at least to family level) have led to their wide use as indicators of ecological health in monitoring practices (Bonada et al. 2006). For this reason, macroinvertebrates have been used in biomonitoring for over a century since the development of the Saprobian system in 1902 (Kolkowitz and Marsson 1902, 1908, 1909).

Diversity indices can provide an indication of ecosystem functioning as there is a general perception that diversity increases with ecosystem health. Two common biodiversity indices are the Shannon Index (Shannon 1948) and

Simpson's diversity (Simpson 1949). These indices provide more information than species richness or abundances alone. Simpson's index relates the number of each species relative to the total number of species at a site. However, this method is heavily weighted to the most abundant species, though is less sensitive to species richness. The Shannon Index is similar but uses log abundances and assumes that individuals are randomly sampled from an independently larger population. These indices do not work on the basis of tolerance/sensitivity and, as diversity does not always relate to ecosystem health, they are criticised for their use in pollution monitoring studies due to their lack of mechanistic bases.

The most well-developed biotic index in the UK is the Walley Hawkes Paisley Trigg index (WHPT) which is a development of the Biological Monitoring Working Party (BWMP) score (Biological Monitoring Working Party 1978). BMWP used expert knowledge to assign taxa with a sensitivity rating to organic pollution between 1 - 10. Organisms with low scores are more tolerant (e.g. 1 =oligochaetes) and organisms with higher scores are more sensitive (e.g. 10 = mayflies and stoneflies). The BMWP score was initially criticised due to misallocation of sensitivity ratings and the potential for error in developing indices based on subjectively derived scores (Walley and Hawkes 1996, Paisley, Trigg, and Walley 2014). The index was later improved through largescale statistical optimisation analysis from abundance data and became the WHPT score which is widely used today, notably in WFD classifications (Walley and Hawkes 1996, 1997, Paisley, Trigg, and Walley 2014). The WHPT index incorporates taxon abundances by adjusting the sensitivity scores for each taxon depending on their log abundance category. Unlike BMWP, WHPT incorporates an abundance measure (log abundance). Using the log abundance, as opposed to the absolute abundance, reflects the semiquantitative nature of the biological sampling method and the potential errors introduced during sorting and identification. The WHPT score for a given site is the sum of all taxon sensitivity scores. However, as this sum is affected by the number of taxa in a sample (WHPT NTAXA), meaning that a more diverse site can have an artificially inflated score, the Average Score Per Taxon (WHPT

ASPT) is then derived. Notably, WHPT only allocates sensitivity at a family level. There can be large differences in sensitivity between species within the same family, as shown in Chironomidae by Zweig and Rabeni (2001). However, the authors also note that species level identification provides limited additional information for the effort required to identify to this resolution, family level is therefore most appropriate. Regardless, the WHPT and related indices are widely used and have been adopted internationally e.g. the BMWP Thailand (Mustow 2002) and the IBMWP Spain (Munné and Prat 2009).

Aquatic ecosystems, especially rivers, are highly dynamic systems strongly influenced by the climate and catchment land-use. This presents challenges when interpreting whether the site-specific conditions are a result of natural variation in water and habitat quality across catchments, or from habitat degradation. The River Invertebrate Prediction and Classification System (RIVPACS) (Wright, Furse, and Moss 1998) seeks to classify river environments based on their characteristics relative to a reference condition (i.e. unaffected by anthropogenic impacts). In order to calculate expected scores, the following environmental variables are entered into the River Invertebrate Classification Tool (RICT): grid reference, altitude, slope, distance from source, discharge category, stream width, stream depth, alkalinity, and substratum characteristics (as percentages of each substrate category - see Table 2.4) (Davy-Bowker et al. 2008). RICT is the interface for RIVPACS which, if ecological data (i.e. taxa abundances) are also provided, can assign an Ecological Quality Ratio (EQR) score which is a ratio of the observed over expected index values (Figure 2.5). Lower EQRs are indicative of sites failing to meet predictions i.e. highly degraded environments. This system has been adopted by water regulation authorities in the UK and the EQRs are used for classification in line with WFD compliance. In the case of WHPT, the lowest score, i.e. the minimum of the NTAXA and ASPT, known as the 'MINTA', is used for WFD classification (Clarke and Davy-Bowker 2014).

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Figure 2.5 – Ecological Quality Ratios and water quality status categories used in Water Framework Directive Classification (from Murray-Bligh 2015).

Pressure-specific indices are those which are optimised using community response to a single stressor. Given the continued degradation of natural environments, these indices can be useful to disentangle the responses in environments which are exposed to multiple-stressors (Berger et al. 2018). Some examples of pressure-specific indices include; the Acid Waters Indicator Community index (AWIC) (Davy-Bowker et al. 2005), the Lotic Index Flow Evaluation (LIFE) (Extence, Balbi, and Chadd 1999) and the SPEcies At Risk index for pesticides (SPEAR) (Liess et al. 2008). Over the last two decades, numerous fine sediment-specific indices have been developed globally (in chronological order of development):

- the Fine Deposited Sediment Biotic Index (DBSI; Zweig and Rabeni 2001)
- the Fine Sediment Biotic Index (FSBI; Relyea, Minshall and Danehy 2012)
- the Combined Fine Sediment Index (CoFSI; Murphy et al. 2015) which collectively represents the organic fine sediment index (oFSI) and the total fine sediment index (ToFSI)
- the PSI group (covered in more detail in Chapter 5);

- The Proportion of Sediment-sensitive Invertebrates (PSI; Extence et al. 2013)
- Empirical Proportion of Sediment-sensitive Invertebrates (EPSI) at species and family level (Turley et al. 2015, 2016)
- the Biological Sediment Tolerance Index (BSTI; Hubler et al. 2016)
- the Multimetric Index (MMI; Doretto et al. 2018)
- the Deposited Fine Sediment Index (DFSI; Gieswein, Hering, and Lorenz 2019)

Historically, most biotic indices have been based on taxonomic approaches which relate species assemblages to environmental conditions. However, the use of functional traits is an emerging concept in ecology. Functional traits are assigned based on the physiological, morphological, ecological and life-history features of an organism (Verberk, van Noordwijk, and Hildrew 2013). Applying functional traits in biomonitoring is based on the theory that traits are filtered according to the prevailing abiotic and biotic conditions (Statzner, Dolédec, and Hugueny 2004). For example, macroinvertebrates (e.g. some Coleoptera) whose eggs can persist in diapause during the dry phase in ephemeral or temporary streams will persist when the sediment is rewetted (Stubbington and Datry 2013). These species can then recolonise quicker than some other taxa whose eggs may not survive desiccation and rely on aerial dispersal. Anthropogenic disturbances (e.g. excessive fine sediment delivery) act as further trait filters which can shape the expected trait composition of macroinvertebrate assemblages according to traits conferring tolerance to the disturbance (Floury et al. 2017). Trait-based approaches have several advantages over traditional approaches. For example, trait-based approaches can transcend boundaries in taxonomic distributions between regions (Lancaster, Downes, and Glaister 2009), they can avoid over emphasis (or under emphasis) of abundant species (Townsend and Hildrew 1994, Verberk, van Noordwijk, and Hildrew 2013), and they can provide a greater mechanistic understanding of the interactions between the environment and ecological community (Doretto et al. 2018).

Many studies have incorporated individual trait responses into quantitative assessments of the effects of fine sediment (e.g. (Rabení, Doisy, and Zweig 2005, Larsen, Pace, and Ormerod 2011, Descloux, Datry, and Usseglio-Polatera 2014, Mathers, Rice, and Wood 2017). The results are sometimes mixed and there is little evidence of unambiguous individual trait responses to fine sediment. Trait-environment relationships can be complex, nonlinear or even characterised by a stress-subsidy response where 'at low stressor levels an ecological variable responds positively until an inflection point beyond which the effect is negative' (Wagenhoff et al. 2012, pii). Consistently strong relationships between individual traits and the environment are rare (Statzner and Bêche 2010) and therefore functional diversity (FD) is often incorporated as an indicator of ecosystem health (Gagic et al. 2015, Schmera et al. 2017). FD is defined as the 'trait variation or multivariate trait differences within a community' (Cadotte, Albert, and Walker 2013, p1080). Buendia et al. (2013) found FD (measured as Rao's quadratic entropy) was sensitive to sediment accumulation. The sediment-specific index PSI was developed using expert knowledge to assign a sensitivity category to each taxon. Therefore, in theory this mechanistic approach should closely link PSI (and therefore EPSI which is an optimisation of PSI) with macroinvertebrate traits. However, there are inconsistences between species scores under this index (and several other indices) and the functional traits possessed by corresponding taxa (Wilkes et al. 2017). Considering these complexities, the utility of trait-based biomonitoring remains unclear.

There have been steps towards directly incorporating trait-based approaches into fine sediment-specific biomonitoring. Murphy et al. (2017) used RLQ analysis to link taxonomic, trait and environmental data and found a limited set of traits through which there was an ambiguous response to fine sediment suggesting potential for incorporation into biomonitoring approaches. Doretto et al. (2018) attempted to develop a biomonitoring tool incorporating some trait-based components. Nevertheless, '*species traits have the potential to disentangle long-term effects of multiple, potentially confounded drivers in ecosystems*' (Floury et al. 2017, p2297). Given the need for effective

management of fine sediment (Mathers et al. 2017), understanding the mechanistic basis for the interactions between the macroinvertebrate assemblage and fine sediment will help to improve these biomonitoring practices.

## 2.5 Conclusion

Excess fine sediment delivery to rivers is a global issue that needs to be tackled by river managers and international research (Mathers et al. 2017). Considering the WFD commitment for every water body to achieve 'good' ecological status by 2027, there is an urgent requirement for targeted monitoring to determine where management methods are required to reduce the delivery of excess fine sediment to aquatic environments (European Community 2000). There is a wealth of evidence quantifying the responses of macroinvertebrates to fine sediment. However, there is conflicting evidence for both taxonomic and functional responses. Thus far, there have been no reviews which are systematic in their nature. A systematic review using a weight-of-evidence approach would help disentangle existing relationships and those which are potentially spurious (Chapter 3). An example of an ambiguous response is the abrasive effects of fine sediments. Further studies are required, i.e. with the use of controlled flume environments and a scanning electron microscope (SEM) following exposure to determine whether soft tissues have been damaged as at present this is just an assumption (Chapter 4). The last decade has seen progress in biomonitoring of fine sediment by either applying basic metrics of community assessment (e.g. abundance and richness), diversity indices, functional trait-based assessments, or by applying biotic indices for general ecosystem health (e.g. WHPT). Several fine sediment-specific biomonitoring indices have been developed for use in the UK. They aim to target the impacts of fine sediment on aquatic communities. However, these indices have been shown to be potentially lacking in their mechanistic links and have yet to be independently tested (Chapter 5).

# Chapter 3 – Fine sediment impacts on aquatic macroinvertebrates: The current state of knowledge

### **Chapter overview**

There have been several narrative literature reviews to date, describing the responses of aquatic organisms to fine sediment. Considering macroinvertebrates significance in biomonitoring practices, and the emergence of sediment-specific biomonitoring tools, the aim of this review was to extract evidence of macroinvertebrate responses to both suspended and deposited fine sediment. Through following a review method adapted from Systematic Maps and Rapid Evidence Assessment, this chapter aims to review the existing literature, quantify the breadth of evidence, analyse the types of responses described, and appraise this through assessment of each selected paper by weighting based on the study design. A total of 8832 articles were extracted from peer-reviewed databases. After the screening process, 131 articles were retained for evidence-based assessment. Using a weight of evidence approach, Chi-squared analysis was used to determine associations between macroinvertebrate responses. Linear modelling was used to determine significant predictors of evidence quality. Results showed a global imbalance of evidence with most research conducted in temperate regions. The majority of evidence was related to articles quantifying deposited, as opposed to suspended sediment. The weight of evidence showed that burrowing organisms were more likely to have a positive response to fine sediment, whereas shredders were more likely to respond negatively. The chapter concludes by making recommendations for future research, highlighting a need to focus on the production of high-quality research with robust study designs focussing on the mechanisms driving macroinvertebrate responses.

This review was conducted with the guidance of Angus Webb (University of Melbourne), co-author of the EcoEvidence review method (Webb et al. 2011). A secondment by M. Mckenzie to the University of Melbourne in April 2016 was funded by the KEEPFISH project (a Horizon2020 Marie Curie RISE project) in order to learn this method of evidence assessment for use in this chapter.

# 3.1 Introduction

## 3.1.1 Methods in evidence review

Synthesising evidence is a key part of the interpretation of existing and emerging information within science. There are numerous ways to review and synthesise scientific evidence. These methods range from traditional narrative literature reviews, which aim to qualitatively describe existing evidence, to Systematic Reviews (SR) which follow a methodical approach and carry out critical appraisals. There is a sliding scale of increased rigour, transparency, time and effort between these two types of review (Figure 3.1) and the different methods which fall between these two ends of this spectrum.

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Figure 3.1 – Evidence review types (from Collins et al. 2015).

Traditional literature reviews are useful tools for synthesising the evidence on a particular subject or theme. However, they can be subjective, lack transparency and repeatability. When searching for evidence, they can introduce publication bias by selecting only highly cited papers or those from a limited group of researchers or journals (Møller and Jennions 2001). Additionally, when synthesising evidence in this way, equal weighting is applied to all studies regardless of their scientific rigour, study design or sample size. This is known as 'vote-counting' (Haddaway et al. 2015). Collectively, these biases can influence the reliability of any conclusions made. Implementing transparency and repeatability through a standardised methodology can help to address this problem (Collaboration for Environmental Evidence 2018). Vote-counting can be overcome by applying a 'weight of evidence' approach. This method assesses the quality of the evidence based on its study design (e.g. replication of factorial design), with higher quality studies providing a greater score or weight in the overall review (Nichols et al. 2011).

Reviews which follow a systematic process aim to reduce biases by following methods which set out the search strategy, evidence recording and assessment (if appropriate). Full SRs represent the most comprehensive form of evidence review. These follow a strict protocol, often governed by expert groups in SRs. For example, The Collaboration for Environmental Evidence, The Evidence for Policy and Practice Information Centre, and The Campbell Collaboration produce guidelines and standards for conducting reviews. The outline of the review, called the review protocol, is peer assessed and the resulting outputs subject to regular updates and monitoring. SRs are labour intensive projects which are exhaustive in their literature search and assessment, and often require input from wide-ranging expertise (Cooke et al. 2017). The prescriptive nature of SRs can be prohibitive and cumbersome which has resulted in a suite of other methods being developed.

Rapid Evidence Assessments (REA) are an emerging method which aim to encompass the rigour and objectivity of a full SR at a fraction of the time and cost. These methods are now being applied to review evidence for policy making decisions. In the context of evidence reviews for use in policy, evidence

can be defined as 'information that can be used to support decisions in developing, implementing and evaluating policy, operations and services' (Collins et al. 2015, piv). The Joint Water Evidence Group produced a method for the production of REAs and quick scoping reviews (QSR) on behalf of the Department of Environment and Rural Affairs (DEFRA) which is now being used in practice (e.g. Water Efficiency and Behaviour Change Rapid Evidence Assessment by Orr, Papadopoulou, and Twigger-Ross 2018). QSRs lie between a standard literature review and an REA (Figure 3.1). They aim to provide an informed conclusion on the volume of evidence in relation to the review question. QSRs generally do not involve any assessment of the robustness or rigour of the evidence and can therefore also lead to votecounting. Systematic maps (SM) also follow the same rigour and objectivity. However, they can be used to address broader questions which are more open and may not have a definitive answer (Berger-Tal et al. 2019). They can also be used to determine knowledge gaps and knowledge clusters (James, Randall, and Haddaway 2016).

Ultimately, the method of review selected will be a result of the requirement for the synthesis of evidence. If the purpose of the review is to place the current topic in context, then a traditional literature review may be sufficient (e.g. Chapter 2 provides an overview of fine sediment and biomonitoring practices which place the rest of this thesis in context). However, if the aim is to identify knowledge gaps and attempt to answer a specific research 'question' from the evidence, then a more structured type of review method would be more appropriate.

### 3.1.2 Reviewing fine sediment effects on macroinvertebrates

In Chapter 2, the wide-ranging potential effects of fine sediment on macroinvertebrates was recognised and discussed (Section 2.3, Figure 2.3). Different components of the macroinvertebrate assemblage may respond to excessive sediment input, depending on their relationship with the substrate, feeding behaviours and other functional traits (Culp, Wrona, and Davies 1986, Angradi 1999, Suren and Jowett 2001, Larsen and Ormerod 2010). There are

both negative and positive responses as some taxa/trait groups benefit whilst others decline or are lost completely. The relationship is not as simple as an inverse association between sediment quantity and macroinvertebrate abundance and is instead a complex web of interactions and effects (Jones et al. 2012b).

To date, several traditional narrative literature reviews have focused on the effects of fine sediment on aquatic organisms, including fish (Kemp et al. 2011), macrophytes (Jones et al. 2012a), macroinvertebrates (Wood and Armitage 1997, Jones et al. 2012b), and diatoms (Jones et al. 2014). As identified in Section 3.1.1, these reviews can be subject to biases. Thus far, there have been no SRs of the literature on this topic, or reviews which have been more systematic in their methodology. Considering the significance of macroinvertebrates in biomonitoring practices (Chapter 2 Section 2.4.2), and the emergence of sediment-specific biomonitoring tools, it is important to develop our understanding of their responses to fine sediment.

A large body of published work exists on the effects of fine sediment on macroinvertebrates, but it remains equivocal and has yet to be quantified using a weight of evidence approach. Investigating these effects lends itself to a method between an SM and an REA. The results of this type of review could lead to a more informed overview of the responses and interactions than the existing traditional literature reviews. Particularly with the emerging use of incorporating traits into biomonitoring tools. It is important to review existing evidence to determine whether any overall conclusions can be made or whether those made in traditional literature reviews are confounded.

### 3.2 Research aims

The aim of this chapter is to evaluate the current state of knowledge on macroinvertebrate responses to excess fine sediment in order to: (1) direct research by identifying key knowledge gaps and; (2) support ongoing efforts to develop effective biomonitoring tools. This will be carried out using a method

adapted from SMs and then critically appraise the evidence extracted using an REA protocol. This review will:

- review existing literature on fine sediment effects on macroinvertebrates
- extract information about each individual study to quantify the breadth of evidence (systematic mapping)
- classify the types of macroinvertebrate responses described (e.g. traditional community indices, trait-based assessments, biomonitoring indices)
- evaluate the causes of macroinvertebrate responses through assessment of the article by weighting evidence based on the study design (i.e. evidence quality)
- assess what factors predict evidence quality
- identify knowledge gaps and make recommendations for future research priorities

# 3.3 Method

## 3.3.1 Review scope

Determining the review scope is an important part of any review. The Population, Intervention/Exposure, Comparator, Outcome (PICO) approach is an established method of breaking down the review scope into its constituent elements (Collins et al. 2015, James, Randall, and Haddaway 2016, Collaboration for Environmental Evidence 2018). The PICO process for this review (Table 3.1) was reviewed by experts in ecology, hydrology, geomorphology, systematic review and policy delivery (Environment Agency, England) at development stage<sup>3</sup>.

<sup>&</sup>lt;sup>3</sup> This review scope was presented to the PhD supervisory team, which comprises the expertise listed, during quarterly supervision meetings

Table 3.1 – PICO (Population, Intervention/Exposure, Comparator and Outcome) analysis of the review scope.

PICO element	Review scope
Population The subject or unit of study	Macroinvertebrates only. Studies including other taxonomic groups will be reviewed but the response on macroinvertebrates must be documented to be extracted as 'evidence'.
Intervention/Exposure The proposed management regime, policy or related intervention/exposure applied or investigated	Exposure to fine sediment (particles <2 mm or sand, silt and clay from land or within channel sources carried in suspension or deposited on and in the river bed)
Comparator The control with no intervention or an alternative to the intervention	Absence of fine sediment; control or reference sites with reduced/enhanced sediment loads; a gradient of fine sediment exposure from low to high.
Outcome The effects of the intervention	Change in macroinvertebrate community structure (i.e. functional or taxonomic indices including biomonitoring index scores) <i>Or</i> Change in population size (e.g. abundance, relative abundance, richness, relative richness) <i>Or</i> Individual behaviour (e.g. drift).

Within the 'Population' element, it is significant to note that the taxonomic group is specifically macroinvertebrates due to their common application in biomonitoring. Additionally, these monitoring practices are only applicable in freshwater lotic systems (streams and rivers), thus any study carried out in marine environments or lentic systems were excluded from this review. The

'Outcome' element is defined as the occurrence and magnitude of a response at the individual and/or community level. It is important to be able to attribute this outcome to fine sediment and not any other confounding effect within each study. Studies which investigate responses in a multi-stressor environment were included as evidence providing the effects of sediment could be isolated from confounding effects. For example, in a full factorial design experiment, only responses recorded from 'sediment only' treatments were included as evidence (as opposed to those crossed with other contaminants) (e.g. Magbanua et al. 2016). As stated in Chapter 2 the effects of sediment associated contaminants and persistent organic pollutants (POPs) are beyond the scope of this thesis. Additionally, the study of the effects of sediment associated contaminants crosses a disciplinary boundary in to the field of ecotoxicology. The mechanisms behind these responses are relatively well understood compared to the effects of fine sediment alone. Furthermore, after initial searches, the wealth of publications relating to specific contaminants was considerably greater (and more than could be reviewed within the capacity of this study) than those investigating the physical effects of sediment alone. Therefore, the scope of this review is therefore limited to the direct physical effects on macroinvertebrates. Fine sediment is most broadly defined as particles <2 mm in diameter (Wood and Armitage 1997, Jones et al. 2012b) so studies investigating the effect of larger size particles (e.g. rock or debris fall) were excluded.

### 3.3.2 Search strategy

Scopus (Elsevier database; see Appendix 1.1 for link to saved search) and Academic Search Complete (EBSCO host) databases were searched on 6<sup>th</sup> October 2018. The use of grey literature (outputs that have not been peerreviewed) has been widely debated; the inclusion of grey literature in systematic reviews is considered an advantage over traditional reviews which only cite peer-reviewed articles (e.g. McAuley et al. 2000). Whilst including grey literature does provide a wider scope than traditional reviews, using peer-reviewed articles gives some assurance that the research is of a certain quality, whereas this may not be the case with grey literature. Additionally, evidence from public
reports or academic theses may be published in part or whole in journal articles, thus risking duplication by including this type of evidence. Grey literature was therefore not included as part of this review.

Finally, it is worth noting that search terms are a critical part of any evidence assessment and must be carefully considered. Due to the broad scope of this review, one search string was used for both databases: 'invertebrates OR macroinvertebrates AND sediment OR fine sediment OR sand OR silt OR clay OR colloid'.

# 3.3.3 Evidence screening

A large number of results were returned from Scopus (7296 articles) and Academic Search Complete (3202 articles). This high number would usually indicate that the search strings need to be revisited and refined because the terms are not specific enough. However, in this instance this result is to be expected given the broad search terms being used and the wide scope of this review. After combining the search results from both databases and removing duplicate publications, the screening was then carried out in a number of phases (Figure 3.2). In screening phase 1, only the title of the publication was assessed and either excluded if irrelevant or taken forward to the subsequent phase. Abstracts were observed for screening phase 2. In the final phase, the full-text articles were assessed for eligibility.

# 3.3.4 Evidence recording

Each piece of evidence equated to a single 'response' (Table 3.2). A 'response' was any evidence within an article of a change to macroinvertebrates, or the macroinvertebrate community, directly attributable to the presence of fine sediment. A single article can yield multiple responses and therefore multiple lines of evidence. Responses were only recorded if they had been achieved using statistical analysis – qualitative observations were not recorded as evidence. Responses were grouped into seven categories (Table 3.2). For the 'subset of population' and 'trait based' response categories, these were further broken down to 'abundance', 'relative abundance', 'richness' and 'relative

richness' depending on how the response was recorded within the article. This avoids double counting as often the same response was recorded across several or all of these subcategories. The 'detectable effect' was then recorded for each response as either positive, negative, mixed (contradictory results within one article) or no effect (no significant difference/effect). To clarify, a 'detectable effect' can be classed as 'no effect' when the evidence presented within an article relates to no significant effect of fine sediment on macroinvertebrates. If the detectable effect was deemed 'indeterminate' (the specific response is listed but it cannot be determined whether the detectable effect is associated with fine sediment or any number of other variables) then the evidence was not recorded.

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Figure 3.2 – Inclusion and exclusion screening process of articles for the REA suggested by PRISMA (Liberati et al. 2009) (n = number of articles).

Table 3.2 – Response categories recorded from each article in evidence assessment.

Category		Description	Subcategories	
1.	Abundance	Total number or density of macroinvertebrates	None	
2.	Taxa richness	Total number of taxa	None	
3.	Diversity	A measure of the diversity of macroinvertebrates, e.g. Shannon's or Simpson's	None	
4.	Subset of population	Any measure of the subset of the population responding to fine sediment, e.g. a single taxa or family.	Categorized by taxonomic resolution Phylum Class Subclass Order Family Subfamily Genus Species	
5.	Biomonitoring index	An index of ecosystem health.	<ul> <li>General index (an index of general stream health e.g. WHPT, BMWP, EPT)</li> <li>Other stressor-specific (an index specific to a particular stressor e.g. LIFE)</li> <li>Sediment-specific (an index specific to fine sediment stress e.g. PSI, CoFSI)</li> </ul>	
6.	Trait based	Functional trait group, an individual trait or preferences of a component	<ul> <li>Aquatic stages</li> <li>Body shape and flexibility</li> <li>Dispersal</li> <li>Feeding mode</li> </ul>	

Category	Description	Subcategories		
6. cont.	of the macroinvertebrate community. The subcategories have been adapted from Tachet et al. (2010).	<ul> <li>Life cycle duration</li> <li>Maximum length</li> <li>Mode of locomotion and relationship to substrate</li> <li>Other</li> <li>Reproduction</li> <li>Reproductive cycles</li> <li>Respiration</li> <li>Substrate preference</li> <li>Type of food</li> </ul>		
7. Other	Any other response e.g. drift	Voltinism     None		
	density or emergence rate.			

For each individual piece of evidence extracted from an article included in the review, the following information was recorded:

- Type of study
  - o Observational
  - Experimental
  - o Both
- Year of article publication
- Country and continent of study location
- Fraction of fine sediment
  - o Deposited
  - Suspended
  - o Both
- Method of sampling macroinvertebrates (quantitative, semi-quantitative or qualitative)
  - Quantitative e.g. Surber or Hess sampler
  - Semi-quantitative e.g. kick net
  - Other e.g. non-standard/qualitative methods

- Method of sampling or measuring fine sediment
  - o Quantitative e.g. cores, or the resuspension method
  - o Semi-quantitative e.g. visual observations
  - Other e.g. non-standard/qualitative methods
- Size of fine sediment (the upper limit of particle size defined within the article) which was also grouped in to one of the following categories:
  - o **≤250 µm**
  - $\circ$  >250  $\mu$ m 500  $\mu$ m
  - $\circ$  >500  $\mu m 1 mm$
  - $\circ$  >1 mm 2 mm
  - **>2 mm**

#### 3.3.5 Assessment criteria

The purpose of assessment criteria is to evaluate the quality of an article and the evidence it contains. Quality assessments were carried out using the EcoEvidence scoring system (Nichols et al. 2011). EcoEvidence is an REA method developed to address causality between environmental stressors, management interventions, and ecological outcomes. The assessment focusses on the study design type and the extent of replication of the reference/control and impact/treatment sampling units to apply a weight of evidence approach. Studies with more robust study designs (e.g. Before-After-Control-Impact) have a higher weight with additional weighting given for a larger number of replicates or sampling units compared to more basic studies (Table 3.3). For experimental mesocosm studies, each individual replicate (providing it isolates fine sediment and not a confounding stressor) was counted towards EcoEvidence scoring, as the nature of mesocosm studies is such that each mesocosm is considered an independent unit. This scoring system is a result of extensive expert consultation and has regularly been used in existing publications (e.g. Vilizzi, Tarkan, and Copp 2015, McInerney et al. 2016, Wilkes, Mckenzie, and Webb 2018)

Table 3.3 – Evidence assessment scoring system (adapted from Norris et al. 2011). The symbols \* and <sup>†</sup> are used to link the study design score with the appropriate replication score (as the replication scoring method is determined by study design). The final EcoEvidence score is calculated as the total of both scores.

Component	Weight			
Study design				
After impact only*	1			
Reference/control vs impact with no before data*	2			
Before vs after with no reference/control location(s)*	2			
Gradient response model <sup>†</sup>	3			
Before After Control Impact (BACI), Before After Reference Impact (BARI), Multiple BACI or beyond BACI*	4			
Replication	4			
*Replication of factorial designs				
Number of reference/control sampling units	0			
0	2			
1	3			
>1				
Number of impact/treatment sampling units				
1	0			
2	2			
>2	3			
<sup>†</sup> Replication of gradient-response models				
<4	0			
4	2			
5	4			
>5	6			

# 3.3.6 Data analysis

In order to understand how the trend in publications on fine sediment has increased over time, Scopus was searched using the terms 'fine sediment'. The number of articles published per year from the period 1966-2017 was extracted. The search term 'Environmental science' was also searched to standardize the results.

A map displaying the number of studies per country was created using the *mapCountryData* function in the *rworldmap* package (South 2016). The data were log transformed prior to mapping to enhance contrast.

In order to determine the strength of association between fine sediment and macroinvertebrate responses, the count data (of responses and detectable effects) were weighted based on the EcoEvidence score for each evidence item. The scores were then divided through the number of individual responses per article within each category. Applying this correction ensures that any associations are based on a weight of evidence approach (i.e. robust studies contribute more to the evidence of a particular interaction) and the results are not biased towards studies which report a large number of responses.

Chi-squared ( $\chi^2$ ) tests were used for testing significant associations between certain 'predictors' or 'responses' and their 'detectable effects' using the weighted count data (Figure 3.3). Chi-squared tests were chosen as the appropriate test for comparing count data to a null (uniform) distribution. Predictors are defined as any of the information recorded for each line of evidence that can influence the outcome of the response and detectable effect (e.g. type of study, sediment size, etc.). Where the predictor or responses were unrelated to the direction (i.e. positive or negative) of the detectable effect, the categories were combined to either 'significant effect' or 'no effect/mixed response'. This is because some taxa would be expected to respond positively and others negatively. For example, when assessing the effect of the sediment size on macroinvertebrates, some taxa would be expected to increase in abundance to an influx of smaller sediment size particles, whereas other taxa would decline.

Chi-squared analysis determines whether the expected cell counts (based on the  $\chi^2$  distribution) significantly vary from the observed cell counts (Scheffé 1947). As there were low observed counts after the weighting procedure, the *simulate.p.values* function was specified during the Chi-squared test. This

function uses 2000 simulations to find the p-value without relying on the Chisquared approximation to the distribution of the test statistic counts. Residuals were plotted using the *corrplot* function (Wei et al. 2017) for every test result that showed a significant association (p <0.05). These plots allow qualitative observations to determine where the associations lie. The size and colour (intensity) of the circles within each plot are indicative of the size of the residuals and therefore the strength of association between each variable. Blue cells specify a positive association between the corresponding row and column, with red cells denoting a negative association. Large bold coloured circles specify a strong association with the circle decreasing in size and colour intensity as the absolute value approaches zero (see Figure 3.7b for an example and an online resource providing further information on the plots is available from sthda 2016).



Figure 3.3 – The classification of information extracted for each line of evidence in the assessment.

In order to determine whether there was any trend between the independent variables within each study and the quality of evidence, linear regression was used. The 'predictors' and 'responses' recorded from each article (see Figure 3.3) were included as independent variables. The country (in which the work was carried out) was aggregated to continent and represented as a categorical variable. The year of publication and the number of responses reported in each

paper were represented as ordinal variables. The remaining variables were binary, e.g. the presence of a measurement of suspended sediment. The assessment of evidence quality (EcoEvidence score) was modelled (using *Im* function in R) against all the information recorded from each piece of evidence offered to the model as independent variables. Model selection was carried out using the *stepAIC* function (direction = 'both') in the MASS package in R (Ripley et al. 2019). All statistical analysis was carried out using R version 3.5 (R Development Core Team 2019).

## 3.4 Results

## 3.4.1 Evidence mapping

The trend in publications citing 'fine sediment' appeared to have followed the trend of 'environmental science' up to the 1990s when, by comparison, it appears to have declined (Figure 3.4). The total number of articles retained for full assessment was 131 (see Appendix 1.2 for a full list). The studies were conducted in 27 different countries (Figure 3.5; Appendix 1.3) with over half of studies coming from only five countries; USA (n = 27), New Zealand (n = 19), UK (n = 15), Canada (n = 12) and Australia (n = 8). A total of 1293 macroinvertebrate responses (i.e. individual items of evidence) were recorded from the 131 articles. The majority of articles (n = 68) consisted of study designs which were 'observational' (i.e. field work sampling one or multiple sites or river reaches), and 58 were 'experimental' (either in-situ such as stream/flow-through mesocosms, or ex-situ and lab-based). Five articles comprised elements of both.

## 3.4.2 Macroinvertebrate responses

The number of responses for each detectable effect and each category can be found in Appendix 1.4. A summary of all Chi-squared tests can be found in Table 3.4 (tables of absolute values of the residuals for each significant test can be found in Appendix 1.5). Results of each test are described in detail below.



Figure 3.4 – Standardized number of publications per year. The standardization was carried out by dividing the number of publications per year citing 'Fine Sediment' over the number citing 'Environmental Science'.



Figure 3.5 – The frequency of articles per country included in the review. Data were log transformed prior to mapping to improve the contrast. Raw values can be found in Appendix 1.3. Countries not filled by colour have zero articles.

Table 3.4 – Results from all Chi-squared tests. Significant results are shown with an asterisk.

Predictor/response		Detectable effects tested		Chi-squared results	
•	Suspended	0	No detectable	p = 0.051*	
•	Deposited		effect or mixed		
•	Both		response		
S	ediment size	0	Detectable effect	$\chi^2 = 45.536$	
•	≤250 µm	0	No detectable	p <0.001*	
•	>250 µm - 500 µm		effect or mixed		
•	>500 µm - 1 mm		response		
•	>1 mm - 2 mm				
•	> 2mm				
Т	aditional metrics	0	No detectable	$\chi^2 = 20.741$	
•	Abundance		effect or mixed	p <0.001*	
•	Richness		response		
•	Diversity (e.g. Shannon's	0	Negative		
	or Simpson's)	0	Positive		
P	opulation subset	0	Detectable effect	Abundance	
•	Phylum/Subphylum	0	No detectable	$\chi^2 = 14.270$	
•	Class		effect or mixed	p = 0.033*	
•	<ul> <li>Subclass</li> </ul>		response	Relative abundance	
•	Order			$\chi^2 = 30.773$	
•	Family			p <0.001*	
•	Subfamily			Richness	
Genus				Not enough data	
•	Species			Deletive vielences	
				Relative richness	
	iomonitoring index		No datastable	Not enough data $y^2 = 24.722$	
D		0	offect or mixed	$\chi^{-} = 21.723$	
				h <0.001	
•			Negativo		
•	Sediment-specific	0	Docitivo		
		0	FUSILIVE		
Tı	ait based: all	0	No detectable	Abundance	
Aquatic stages			effect or mixed	$\chi^2 = 65.910$	
			response	p <0.001*	

Predictor/response	Detectable effects tested	Chi-squared results	
<ul> <li>Body shape and flexibility</li> <li>Dispersal</li> <li>Feeding mode</li> <li>Life cycle duration</li> <li>Maximum length</li> <li>Mode of locomotion and relationship to substrate</li> <li>Other</li> <li>Reproduction</li> <li>Reproductive cycles</li> <li>Respiration</li> <li>Substrate preference</li> <li>Type of food</li> <li>Voltinism</li> </ul>	<ul> <li>Negative</li> <li>Positive</li> <li>Detectable effect</li> <li>No detectable effect or mixed response</li> </ul>	Relative abundance $\chi^2 = 34.214$ $p = 0.001^*$ Richness $\chi^2 = 6.609$ $p < 0.001^*$ Relative richness           Not enough data           Abundance $\chi^2 = 23.579$ $p = 0.003^*$ Relative abundance $\chi^2 = 23.534$ $p < 0.001^*$ Richness $\chi^2 = 19.589$ $p < 0.001^*$ Relative richness           Not enough data	
Trait based: Feeding mode <ul> <li>Collector</li> <li>Filterer</li> <li>Other</li> <li>Predator</li> <li>Scraper</li> <li>Shredder</li> </ul>	<ul> <li>No detectable effect or mixed response</li> <li>Negative</li> <li>Positive</li> </ul>	Abundance $\chi^2 = 28.694$ $p < 0.001^*$ Relative abundance $\chi^2 = 21.170$ $p = 0.006^*$ Richness           Not enough data           Relative richness $\chi^2 = 22.825$ $p < 0.001^*$	
<ul> <li>I rait based: Mode of</li> <li>locomotion and</li> <li>relationship to substrate</li> <li>Burrower</li> <li>Climber</li> </ul>	<ul> <li>No detectable effect or mixed response</li> <li>Negative</li> <li>Positive</li> </ul>	Abundance $\chi^2 = 50.912$ p <0.001* <b>Relative abundance</b> $\chi^2 = 67.441$ p <0.001*	

Predictor/response		Detectable effects tested	Chi-squared results	
•	Clinger		Richness	
•	Crawler		Not enough data	
•	Depositional			
•	Erosional		Relative richness	
•	Sprawler		Not enough data	
•	Swimmer			

The majority of evidence was related to sediment in the deposited fraction (articles = 85, responses = 949) compared to sediment in the suspended fraction (articles = 26, responses = 167), however 20 articles and 177 responses measured both fractions (Figure 3.6). The fraction of fine sediment recorded across all articles ranged from 63  $\mu$ m – 4 mm. The sediment size was not significantly associated with the detectable effect of the macroinvertebrate responses ( $\chi^2$  = 5.661, p = 0.060).



Figure 3.6 - Number of articles and responses recorded per sediment fraction described.

The particle size definition of fine sediment differed considerably between articles (Figure 3.7a). A large number of articles (n = 30) did not specify particle size. Chi-squared analysis showed that sediment size had a significant association with detectable macroinvertebrate responses ( $\chi^2$  = 45.536, p <0.001). There was a strong positive association between the weight of evidence in studies defining particle size as >1 – <2 mm and the likelihood of finding a detectable effect on macroinvertebrates (Figure 3.7b, Table A1.4 Appendix 1.5). Studies defining fine sediment particle size as ≥2 mm, were more likely to report a mixed response or no significant detectable effect.

The 'population subset' was the most frequently presented response category with 437 total responses, followed by 'traits or trait-based indices' with 319 responses. This is a result of articles presenting multiple lines of responses under this category, for example (Lange, Townsend, and Matthaei 2014) presented 69 responses which fit within this category. The taxonomic resolution of the responses recorded in this category varied across eight taxonomic levels, with family, genus and order the most common (Figure 3.8a). Chi-squared analysis showed that taxonomic level was significantly associated with a detectable effect on macroinvertebrates when the subset was recorded as abundance (Figure 3.8b;  $\chi^2 = 14.270$ , p = 0.030) and relative abundance (Figure 3.8c;  $\chi^2 = 30.773$ , p <0.001).

For responses quantified as absolute abundance, there was a strong positive association between the taxonomic levels class, subfamily and genus and a significant detectable effect of fine sediment but at order and subclass levels the association was negative (Figure 3.8b; Table A1.5 Appendix 1.5). For responses quantified as relative abundance, the only notable association was a negative relationship between taxonomic level family and a significant detectable effect of fine sediment (Figure 3.8c; Table A1.6 Appendix 1.5). At species level, the highest taxonomic resolution, there was no particularly strong association for either detectable effects for both absolute and relative abundance.



Figure 3.7 – Frequency of particle size reported from articles included in evidence review (a) and residual plots from the Chi-squared test between the particle size range and the detectable effect from fine sediment using weight of evidence (b). The size and colour of the circles within each plot are indicative of the size of the residuals and therefore the strength of association between each variable. The colour ramp shows the size and sign of residual. Therefore, blue cells specify a positive association between the corresponding row and column, with red cells denoting a negative association. Large and bold coloured circles specify a strong association with the circle decreasing in size and colour intensity as the absolute value of the residual approaches zero.



Figure 3.8 - Frequency of responses for each taxonomic level from articles included in the evidence review (a) residual plots from the Chi-squared tests between the taxonomic level and the detectable effect from fine sediment where the subset of the population was recorded as an abundance (b) and relative abundance (c). The size and colour of the circles within each plot are indicative of the size of the residuals and therefore the strength of association between each variable. The colour ramp shows the size and sign of residual. Therefore, blue cells specify a positive association between the corresponding row and column, with red cells denoting a negative association. Large and bold coloured circles specify a strong association with the circle decreasing in size and colour intensity as the absolute value of the residual approaches zero.

Most articles presented information on traditional metrics: abundance, richness and diversity index (Figure 3.9a). Chi-squared analysis showed that there was a significant association between the detectable effect of fine sediment and the traditional metrics ( $\chi^2 = 20.741$ , p = 0.001, Table A1.7 Appendix 1.5). There was a strong positive association between abundance and a positive detectable effect (i.e. increased fine sediment increases abundance). However, the opposite was true for richness and diversity (i.e. increases in fine sediment reduce richness and diversity).



Figure 3.9 - Frequency of responses for abundance, richness and diversity (a) and plot of residuals from the Chi-squared test between these traditional metrics and the detectable effect from fine sediment. The size and colour of the circles within each plot are indicative of the size of the residuals and therefore the strength of association between each variable. The colour ramp shows the size and sign of residual. Therefore, blue cells specify a positive association between the corresponding row and column, with red cells denoting a negative association. Large and bold coloured circles specify a strong association with the circle decreasing in size and colour intensity as the absolute value of the residual approaches zero.

The majority of trait-based responses were related to functional feeding groups (Figure 3.10a). Chi-squared analysis showed that functional feeding group was significantly associated with the detectable effect when the feeding group was recorded as an abundance ( $\chi^2 = 29.327$ , p = 0.004) and relative abundance ( $\chi^2$ 

= 26.233, p = 0.003). For abundance, there was a strong positive association between collectors and mixed or no detectable effect (Figure 3.10b; Table A1.8 Appendix 1.5). There was a strong positive association between shredders and a negative detectable effect (i.e. increased fine sediment decreases abundance of shredders). When the response was recorded as relative abundance (Figure 3.10c; Table A1.9 Appendix 1.5), the was a strong positive association between collectors and filterers have a positive detectable effect.

Chi-squared analysis showed that the locomotion mode and relationship to substrate trait group was significantly associated with a detectable effect when the response was recorded as abundance ( $\chi^2 = 50.912$ , p <0.001) or relative abundance ( $\chi^2 = 67.441$  p <0.001). There was a strong positive association between burrowers and a positive detectable effect (Figure 3.11a; Table A1.10 Appendix 1.5) (i.e. increased fine sediment increases abundance of burrowers). There was a moderate association between clingers and a negative detectable effect.

When the response was recorded as relative abundance, burrowers were associated with a positive detectable effect (Figure 3.11b; Table A1.11 Appendix 1.5). There was a moderate negative association between burrowers and a negative, mixed response or no detectable effect. There was a strong positive association between both clingers and organisms living in erosional habitat types and a negative detectable effect. Only trait modalities with sufficient data are presented in residual plots.

The weight of evidence from studies that reported biomonitoring indices was significantly associated with a detectable effect on macroinvertebrates ( $\chi^2 = 21.723$ , p <0.001). Seven studies presented results on sediment-specific indices: all of the responses showed a negative effect. For sediment-specific and other stressor-specific indices, there was a strong negative association with a mixed/no effect (i.e. these indices were highly unlikely to have a mixed or no effect) and a moderate positive association with a negative effect (Figure 3.12; Table A1.12 Appendix 1.5). The general indices showed no strong associations with any detectable effects.



Figure 3.10 - Frequency of responses for each trait category (a) and plots of the residuals from the Chi-squared test between each functional feeding group and the effect, where the functional feeding group was recorded as abundance (b) and relative abundance (c). The size and colour of the circles within each plot are indicative of the size of the residuals and therefore the strength of association between each variable. The colour ramp shows the size and sign of residual. Therefore, blue cells specify a positive association between the corresponding row and column, with red cells denoting a negative association. Large and bold coloured circles specify a strong association with the circle decreasing in size and colour intensity as the absolute value of the residual approaches zero.



Figure 3.11 - Plot of the residuals from the Chi-squared test between the trait group 'mode of locomotion' and 'relationship to substrate' and the detectable effect to fine sediment where the trait modality was recorded as abundance (a) and relative abundance (b). The size and colour of the circles within each plot are indicative of the size of the residuals and therefore the strength of association between each variable. The colour ramp shows the size and sign of residual. Therefore, blue cells specify a positive association between the corresponding row and column, with red cells denoting a negative association. Large and bold coloured circles specify a strong association with the circle decreasing in size and colour intensity as the absolute value of the residual approaches zero.

# 3.4.3 Predictors of evidence quality

The linear model predicting evidence quality from the predictor variables was significant, however it explained a low amount of total variance (Adj R<sup>2</sup>=0.237, F=4.368, p=<0.001; diagnostic plots Appendix 1.6). The presence of suspended sediment measurements, the reporting of abundance/density responses, and observational study types were significantly predictive of EcoEvidence score (Table 3.5). Studies which report these variables were likely to have lower EcoEvidence scores (i.e. of lower quality). Biomonitoring index and other methods of measuring fine sediment (see Section 3.3.5) were significant

positive coefficients-estimates. Therefore, studies which report these variables were likely to have higher EcoEvidence scores (i.e. be of higher quality).



Figure 3.12 – Plot of residuals from the Chi-squared test between biomonitoring indices and the detectable effects. General indices aim to detect a range of pollutants, sediment-specific indices are exclusive to fine sediment, and other stressor specific are related to another specific pollutant (e.g. flow and the LIFE index). The size and colour of the circles within each plot are indicative of the size of the residuals and therefore the strength of association between each variable. The colour ramp shows the size and sign of residual. Therefore, blue cells specify a positive association between the corresponding row and column, with red cells denoting a negative association. Large and bold coloured circles specify a strong association with the circle decreasing in size and colour intensity as the absolute value of the residual approaches zero.

# 3.5 Discussion

The aim of this chapter was to map and review the existing evidence which quantifies the effects of fine sediment on macroinvertebrates. Although there have been several traditional literature reviews published on the effects of fine sediment on macroinvertebrates (e.g. Wood and Armitage 1997; Jones et al. 2011), this review is the first of its kind that follows a methodological approach which encompasses the rigour and objectivity of a systematic review. The majority of the literature relating to ecological responses cited within the discussion has been restricted to the articles which were assessed as part of this evidence review (see Appendix 1.2 for a complete list). This is in order to focus the discussions of the results based around the evidence included in the review and avoid duplication of a broader, more narrative review style (such as Chapter 2). Additional literature has been used to support arguments and is cited as appropriate.

Table 3.5 – Summary results from the general linear model determining predictors of overall evidence quality (assessed using EcoEvidence). Significant coefficients are indicated with an asterisk.

Coefficients	Estimate	Std.	z value	р
		Error		
Intercept	10.632	2.535	4.195	<0.001*
Suspended sediment	-1.312	0.461	-2.850	0.005*
Continent – Asia	-0.411	2.057	-0.200	0.842
Continent – Europe	2.117	1.798	1.178	0.241
Continent – North America	1.720	1.786	0.963	0.338
Continent – Oceania	2.469	1.819	1.357	0.177
Continent – South America	3.250	1.958	1.660	0.100
Study type – Observational	-1.845	0.506	-3.650	<0.001*
Response type –	-1.608	0.480	-3.347	0.001*
Abundance/density				
Response type - Biomonitoring index	1.526	0.516	2.959	0.004*
Method of measuring	-4.236	3.043	-1.392	0.167
macroinvertebrates – Quantitative				
Method of measuring	-4.893	2.981	-1.642	0.103
macroinvertebrates – Semi-				
quantitative				
Method of measuring fine	2.634	1.177	2.239	0.027*
sediment – Other				

#### 3.5.1 Breadth of evidence

Despite the evidence search yielding a large number of results (8832 unique articles from two databases), only 131 articles were suitable for inclusion in the review. The main reasons articles were excluded from the review were because fine sediment either was not quantified or the effects were impossible to separate from the interactions of other environmental variables. Many of these studies were more generalised studies looking at variation in environmental gradients between stream sites/reaches, rather than focussing on fine sediment effects. A large number of studies included in the review presented gradient responses: they sampled a number of rivers or reaches over a gradient of suspended and/or deposited fine sediment to determine, often though multiple regression analysis, if fine sediment was a significant predictor of macroinvertebrate community composition (e.g. Angradi 1999; Braccia and Voshell 2006). In dynamic river systems, there are often other significant factors affecting macroinvertebrate responses such as flow (Ehrhart, Shannon, and Jarrett 2002, Fritz, Dodds, and Pontius 2009, Espa et al. 2015) which can be interlinked with the effects of fine sediment.

The results of the assessment of the number of studies published citing 'fine sediment' followed the trend for increasing numbers of publications over time. This search term would have captured results from all scientific fields including ecology, hydrology and geomorphology. Despite the evidence that the excessive erosion, transportation and deposition of fine sediment is now recognised as a major threat to ecosystems globally, particularly within the last 20 years (Wood and Armitage 1997, Owens et al. 2005), it appears there has been a decline in published studies relative to all environmental science literature. Speculatively, this decline in publications relative to the broader scientific field could be because research efforts have focused on other issues such as emerging pollutants (e.g. microplastics) or climate change.

The majority of articles included in the review were conducted in temperate regions, with most studies originating from only four countries (USA, New Zealand, UK and Canada). Studies based in equatorial or tropical zones were

lacking. There is potential for this skew to be an artefact of search strategy as only evidence published in English was included in the review. However, Schmera et al. (2017) also found a global bias towards Europe and North America when conducting an evidence review on functional diversity. This geographical skew indicates a deficiency of evidence on the effects in these regions where the climate presents unique hydrological and geomorphological conditions. Considering the rate of change of land use in countries such as Brazil, Indonesia and Malayasia, fine sediment erosion from land poses a significant threat (Ponsioen and Blonk 2012). Further study in a broader geographical range could present new evidence and potentially novel responses of the effects of excessive fine sediment.

The particle size definition of fine sediment varied considerably among the studies reviewed. Thirty studies failed to specify particle size definition despite being explicit about studying the effects of fine sediment. Some studies quantified suspended sediment concentration (SSC) through the use of turbidity or directly measured concentration but did not specify the size range (e.g. Miliša, Živković, and Habdija 2010; Culp et al. 2013; Blettler et al. 2015). The size range of sediment carried in suspension can vary according to the hydraulic conditions (Van Rijn 1993). Fine sediment is generally considered to compose the fractions sand, silt and clay. The Wentworth scale (Wentworth 1922) classifies the particle size of sand as 0.0625-2 mm, silt 0.0039-0.0625 mm and clay <0.0039 mm. However, there is little agreement in the evidence, with definitions of particle size ranging from <63  $\mu$ m to 4 mm (fine gravel). This implies that there is no international definition of fine sediment particle size, making comparisons between studies problematic. The specific particle size distribution is thought to have distinctive impacts on the response of the aquatic community (Richards and Bacon 1994). Davis et al. (2015) sampled multiple sites on a single river and found that average sediment size was a significant predictor of 11 out of 25 macroinvertebrate taxonomic responses reported. Duan, Wang, and Tian (2008) used colonisation experiments and found that a number of responses, including taxonomic richness, density, and diversity, were lowest in the finest particle size chamber compared to the coarsest (median

diameter 0.2 mm compared to 1.5 mm). However, there is conflicting evidence. Bond and Downes (2003) added sediment in the particle size range 0.5 - 1 mm to experimental channels but saw no significant effect on the abundance or species richness in the drift. Whereas, Culp, Wrona, and Davies (1986) added sediment of this same specific size range to riffles and saw a significant change in both macroinvertebrate density and drift response. When considering the weight of evidence across all responses, particle size in the range 1 - 2 mm was most likely to have a significant effect on macroinvertebrates. Considering the number of articles which record fine sediment as particles <2 mm (56 articles), and given Wentworth's size classification for sand, silt and clay, the most acceptable definition of fine sediment is to include all particles <2 mm.

Most evidence was related to sediment in the deposited fraction. Deposited sediment was measured in various ways, with visual estimates (percentage cover) being the most common. Studies that observed sediment in the suspended fraction predominantly focussed on sediment flushing from dams (Espa et al. 2015) or increased sediment loads from construction (Fossati et al. 2001, Ehrhart, Shannon, and Jarrett 2002, Couceiro et al. 2010a). Although most studies quantifying suspended sediment were observational (68 articles), a substantial number of studies were experimental (58 articles) and 2 articles contained elements of both (Doeg and Milledge 1991, Bond and Downes 2003, de Castro Vasconcelos and Melo 2008). In a field experiment, Culp, Wrona, and Davies (1986) compared the effects of both suspended and deposited sediment by adding sediment to either transporting or depositing riffles with existing similar communities. After treatment, the transporting riffle had significantly lower total macroinvertebrate density. The fraction of fine sediment was not significantly associated with the effect on macroinvertebrates (from Chi-squared analysis). However, studies which measured suspended sediment had significantly lower EcoEvidence (evidence quality) scores. This is typically because the large majority of articles which reports suspended sediment are related to studying the effects of dam flushing or construction and therefore often only a single river is sampled.

#### 3.5.2 Evidence of macroinvertebrate responses to fine sediment

The majority of macroinvertebrate responses fit within the 'population subset' category, i.e. a change in abundance, relative abundance, richness or relative richness of a specific species, genus or other taxonomic rank. Taxonomic resolution of the population subset category was significantly associated with an effect on macroinvertebrates (i.e. taxonomic resolution affected whether there was a significant macroinvertebrate response or not). Identification at the species level, for responses recorded as abundances, did not have a particularly strong association with detectable effects on macroinvertebrates. The reason for this is not immediately obvious. At the highest taxonomic resolution, there are expected to be large variabilities in sensitivities between species. Sensitive species would be expected to show a negative response. whereas tolerant species would be expected to a positive response or potentially no effect (i.e. there is no adverse effect from fine sediment so can indicate tolerance). By comparison, to show a significant response at the family level responses, only a single sensitive or tolerant taxon would be sufficient to drive the overall effect.

There was a positive association between genus and subfamily and a significant effect when the response was recorded as absolute abundance. This is because of the high number of articles presenting results on genera within the Ephemeroptera, Plecoptera and Trichoptera (EPT) orders. These orders are typically sensitive to pollution and the EPT index (calculated as the number of EPT families within a single sample) is used as a generalised biomonitoring index. For example, Gomi et al. (2010) and Buendia et al. (2013) presented evidence from numerous genera within the EPT orders that all responded negatively to increasing fine sediment. This is partially supported by the analysis for taxonomic levels presented as relative abundance. However, family is the only taxonomic level which shows a strong association when the rest of the taxonomic community is considered (relative abundance).

There were many evidence items related to Chironomidae (Diptera). However, there was conflicting evidence for this taxon. Twenty-six evidence items

presented evidence at the family level. Seventeen evidence items showed no effect or a mixed response, six items showed a negative response and three items showed a positive response. This can be partly explained by the higher level of taxonomic resolution used. In the UK, this taxon is commonly identified to the subfamily level for water monitoring purposes. Conflicting evidence is similarly apparent at the subfamily level. For example, Tanypodinae responded negatively to suspended sediment in Fossati et al. (2001) yet it exhibited a positive response to deposited sediment in two studies by Piggott et al. (2012) and Piggott, Townsend, and Matthaei (2015). Orthocladiinae responded negatively to suspended sediment in Gray and Ward (1982), Fossati et al. (2001), and to deposited sediment in Descloux, Datry, and Marmonier (2013). Although, it was shown to respond positively by Magierowski et al. (2015). There is a tendency for chironomids to be considered as tolerant to harsh conditions, however the evidence points to substantial variability at taxonomic resolutions finer than family level. Chironomidae larvae are considered one of the most abundant and diverse freshwater macroinvertebrate taxa with over 1200 species in Europe, inhabiting all types of permanent and temporary water bodies (Spies and Saether 2004, DeWalt, Resh, and Hilsenhoff 2010). The variation in life-history strategies and traits has resulted in the use of Chironomidae as indicators of anthropogenic disturbance (Serra et al. 2017).

Analysis showed that studies reporting traditional indices (e.g. Mary and Marmonier 2000; Fritz, Dodds, and Pontius 2009) were more likely to report a detectable effect on macroinvertebrates. The analysis showed that abundances were likely to increase with fine sediment. This is because small bodied species which colonise quickly and persist in high numbers are more likely to be tolerant to fine sediment and persist and thrive in environments of poor quality. The results also showed that richness and diversity were less likely to respond positively to fine sediment, which supports existing ecological theories of effects from disturbance (Connell 1978).

Functional feeding group was the most common trait response reported in the literature reviewed. An increase in fine sediment deposition can bury food resources (Couceiro et al. 2010b), affect quality and quantity of periphyton

(Buendia et al. 2013a), reduce exchange of water and dissolved substances (i.e. hyporheic exchange flow) (Descloux, Datry, and Usseglio-Polatera 2014) and dilute available food resources (Broekhuizen, Parkyn, and Miller 2001). The results of the evidence review showed that filterers were more likely to have a positive response to fine sediment. This is in conflict with previous traditional reviews (e.g. Jones et al. 2012b) which generalise filter feeders as sensitive to fine sediment. However, this interaction could be part of a subsidy-stress response where an initial increase in fine sediment could increase food supply and be beneficial for filter feeders. In clear water conditions, there is low supply of fine particulate organic matter in suspension. With adequate supply of organic particulates, filter feeding becomes viable. As fine sediment continues to increase further, filter feeding becomes ineffective as feeding apparatus becomes clogged and gut filling by inorganic particles occurs (Lemly 1982, Strand and Merritt 1997, Fossati et al. 2001). Collectors were also more likely to have a positive response to fine sediment. Collectors (or gatherers/collectorgatherers) are thought to substitute shredders in impacted sites and are generally present regardless of water or habitat quality (Couceiro et al. 2010b). It was clear from the results that shredders are most likely to have a negative response to fine sediment. Therefore, shredders are likely to be the most sensitive trait group. This is consistent with the findings of Wilkes et al. (2017) which found shredders to be consistently associated with sensitivity scores across five fine sediment-specific indices. This is the most unequivocal trait-fine sediment relationship (e.g. Rabení, Doisy and Zweig 2005; Scott and Zhang 2012; Mathers, Rice, and Wood 2017). The mechanisms behind shredder sensitivity are thought to be associated with burial of leaf litter and a reduction in its quality through inhibition of fungal growth (Couceiro et al. 2010b, Doretto et al. 2016, Louhi, Richardson, and Muotka 2017).

The weight of evidence showed burrowers were more likely to have a positive response to fine sediment. Increased fine sediment deposited on the stream bed will provide additional habitat for burrowers (Griffith et al. 2009). Additionally, burrowers living within the interstices can be adapted to lower oxygen conditions. However, burrowers of coarser substrates (e.g.

Ephemeridae) can be sensitive to fine sediments (Wilkes et al. 2017), therefore this trait response is more ambiguous. When considering relative abundance, taxa inhabiting erosional areas were more likely to have a negative response. This is intuitive as erosional areas will transport sediment, and taxa living in these areas will be less tolerant of sediment depositing areas.

There was no association between indices for general health and detectable effects on macroinvertebrates. This is partly because an increase or decrease in an index score does not equate the same meaning for all these general indices. Jun et al. (2011) and Phillips et al. (2016) presented variations of the dominance index. As fine sediment increases and a smaller number of tolerant taxa persist and become dominant, this index would increase. Whereas the EPT index, which was commonly presented throughout the evidence, would decrease in response to increased fine sediment as these taxa are sensitive to fine sediment (e.g. Angradi 1999; Espa et al. 2013; Piggott, Townsend and Matthaei 2015). However, EPT showed no effect or a mixed response in twenty evidence items and even a positive response in two evidence items (Ramezani et al. 2014, Phillips et al. 2016). Employing blanket sediment guidelines is considered a poor way to manage and monitor fine sediment because of varying catchment conditions and natural background levels of fine sediments (Foster et al. 2011, Collins et al. 2012, Bilotta et al. 2012). Therefore, understanding the mechanisms that fine sediment affects communities is crucial to the development, improvement and adoption of biological monitoring practices for fine sediment.

Stressor-specific or sediment-specific indices unequivocally showed negative responses to fine sediment. This shows the value of stressor-specific biomonitoring indices compared to those which aim to assess only general ecosystem health. Only seven articles presented evidence of a sediment-specific biomonitoring index response. All these articles were from research in the UK using UK developed fine sediment biomonitoring indices. This reflects the recent development of sediment-specific tools. For example, the Proportion of Sediment-sensitive Invertebrates, PSI, is a sediment-specific biomonitoring index that was developed for application in the UK in 2011 (Extence et al.

2013). This index was refined with empirical weightings in 2015 (EPSI) (Turley et al. 2015, 2016). International developments of sediment-specific fine sediment biomonitoring indices include the Fine Deposited Sediment Biotic Index (DBSI) (Zweig and Rabeni 2001), the Fine Sediment Biotic Index (FSBI) (Relyea, Minshall, and Danehy 2012). However, all of these articles were removed at the screening stage as the metric of fine sediment was not quantified. Studies which presented biomonitoring index results were more likely to have higher EcoEvidence scores (evidence quality). This is because when presenting these indices, they are often calibrated with a large number of sites, therefore scoring more highly under EcoEvidence (Table 3.3).

The objective of this study was to collate the evidence on macroinvertebrate responses to fine sediment. When extracting the evidence, it became apparent that there was little evidence on the specific mechanisms which drive the effects of the responses. Mechanisms are clearer from the outcomes of experimental studies. For example, Broekhuizen, Parkyn, and Miller (2001) found that increasing the sediment to food ratio reduced assimilation rates in direct proportion to the sediment fraction added in grazers. Albertson and Daniels (2016) studied the effects of *Hydropsyche* sp. feeding rate from fine sediment effects on feeding net architecture. Some articles attempted to suggest or hypothesise mechanisms through which fine sediment is driving the change, for example through habitat alteration (Braccia and Voshell 2006, Chakona et al. 2009, Gomi et al. 2010), effects on oxygen availability and hyporheic exchange (Descloux, Datry, and Usseglio-Polatera 2014), or food availability (Fossati et al. 2001, Blettler et al. 2015). Several articles discuss the idea of abrasion to soft or exposed tissues and feeding structures as a driving factor (Culp, Wrona, and Davies 1986, Bond and Downes 2003, Chiu et al. 2013). However, there was no quantitative evidence that confirms abrasion effects. The majority of articles reviewed did not suggest or mention the mechanisms driving the response. Researchers have highlighted that although they could quantify the responses using quantitative data, the direct and indirect effects and mechanisms driving this change could not be disentangled (Connolly and

Pearson 2007, Cover et al. 2008, Buendia et al. 2013b, Culp et al. 2013) and that further research on mechanisms is needed (Conroy et al. 2016a).

## 3.6 Conclusions

There is a wealth of publications containing quantitative evidence of the responses of macroinvertebrates to fine sediment. A range of traditional reviews have used this existing evidence to describe the effects. However, these traditional literature reviews could be susceptible to biases such as an overemphasis of poor-quality studies. For example, 'abrasion' and 'clogging' of macroinvertebrate body parts by fine sediment is widely cited (e.g. Wood and Armitage 1997, Jones et al. 2012b) yet unsubstantiated by direct evidence. The review presented here provides the first methodological and systematic type approach to assessing the responses of macroinvertebrates to fine sediment. This review has identified knowledge gaps in equatorial regions which could provide insights to novel macroinvertebrate responses. Additionally, the range of definitions of sediment particle size across all publications imply there is no global definition which creates problems when making comparisons across evidence. Using a weight of evidence approach, the results show that shredders are unequivocally sensitive to fine sediment and burrowers appear tolerant. In the case of burrowers, this result should be observed with caution as the size of burrowing material is an important factor in determining the sensitivity of taxa in this trait group. Finally, the results of this review support the continued development of sediment and other stressor specific biomonitoring indices through the production of high-quality research.

There needs to be a focus on the production of high-quality research with robust study designs incorporating replication of both treatment/reference and control/impact units. In this review, a large number of studies (8832) were extracted from the literature searches yet only a small proportion were included in the final review. After the weighting approach, there was insufficient data to statistically analyse some of the interactions. More research needs to be

conducted in tropical and equatorial areas to redress the balance with those carried out in temperate zones. The use of biomonitoring indices when quantifying the effects of fine sediment shows consistent results but there was a relatively small number of articles using these tools. This implies that their use is still relatively early in adoption and there is potential for these to be developed to ensure they are based on the latest evidence and as we continue to develop our understanding of macroinvertebrate responses to fine sediment. There is some consistency in the evidence of trait responses, particularly shredders, to fine sediment which could be incorporated into future monitoring practices.

The understanding of mechanisms driving the responses to fine sediment needs to be developed. By achieving this, direct causal links can be identified as opposed to merely observing correlations and patterns. This could be achieved through a greater focus on controlled experimental studies rather than large scale, multi-catchment gradient studies. For example, an exposure flume experiment to quantify how suspended sediment causes abrasion to individual species which could explain invertebrate drift responses in natural conditions (Chapter 4). Understanding the mechanisms by which fine sediment affects macroinvertebrates is important to improve and develop monitoring methods for fine sediment globally.

# Chapter 4 – Physical effects of suspended fine sediment on lotic macroinvertebrates

## **Chapter overview**

Building on knowledge gaps identified in Chapter 3, this chapter investigates the potential for physical damage caused by suspended fine sediment on gills of three macroinvertebrate species, Hydropsyche siltalai, Ephemera danica and Ecdyonurus venosus. Macroinvertebrate cadavers were exposed to three suspended sediment concentrations (SSC) (control 3.5, low 83.7 and high 404.0 mg  $l^{-1}$ ) at two velocities (low 0.19 m s<sup>-1</sup> and high 0.37 m s<sup>-1</sup>), for six hours in a recirculating flume. Tracheal gill surfaces were examined for signs of physical damage using Scanning Electron Microscopy (SEM) images. Physical damage predominantly consisted of fine sediment coverage of gill surfaces, appearing as a deposited layer of sediment obscuring and potentially clogging the gill. For *E. venosus*, SSC influenced gill coverage but velocity had no significant effect. Coverage of *H. siltalai* gill surfaces increased significantly between low and high SSC but only at the higher flow velocity. Finally, E. danica gill coverage did not differ significantly across any concentration. Variation in gill structure and function between the three species, as well as their habitat preferences, can help explain the results. There was limited evidence of abrasion as a direct physical effect of fine sediment, in contrast to its widely cited occurrence in the literature.

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#### 4.1 Introduction

Macroinvertebrate responses to fine sediment represent a complex mix of direct and indirect effects. In Chapter 3, the large body of evidence of macroinvertebrate responses to fine sediment was reviewed. There is a wealth of evidence which quantifies community level responses to fine sediment, such as changes in abundance (Blettler et al. 2015, Elbrecht et al. 2016), functional traits (Rabení, Doisy, and Zweig 2005, Larsen, Pace, and Ormerod 2011, Wagenhoff, Townsend, and Matthaei 2012) or biomonitoring index scores (Jun et al. 2011, Glendell et al. 2014, Conroy et al. 2016a). Research has also quantified the effects of suspended sediment on feeding efficiency (Kefford et al. 2010), egg survival (Everall et al. 2018), and the effect of burial by sediment deposition (Wood et al. 2005, Conroy et al. 2018). However, research which considers the direct physical effects of fine sediment in suspension at the organism level is limited. Based on existing evidence, there are likely to be two main processes through which suspended sediment affects macroinvertebrates physically: (i) coverage of fine sediment on tissues and external structures, potentially leading to clogging effects; and (ii) abrasion - physical damage in the form of scrapes or scratches from the angularity of fine sediment particles in suspension or saltation.

Clogging effects from fine sediment were first defined by Lemly (1982) as the accumulation of particles on body surfaces and respiratory structures. These effects have been reported in fish, affecting gaseous exchange through the gill epithelium and disrupting respiration (Cordone and Kelley 1961, Bond and Downes 2003) and osmoregulation (Bruton 1985, Waters 1995, Bergstedt and Bergersen 1997). Similarly, for macroinvertebrates, fine sediment can also build-up on external organ surfaces and disrupt the normal functioning of gills and filter-feeding apparatus (Strand and Merritt 1997, Allan 2004), however, the evidence remains uncertain. The rationale linking the effects of fine sediment to clogging predominantly concerns filter feeders that may spend extra time cleaning feeding structures (e.g. Cladocera - Arruda, Marzolf, and Faulk 1983; Hart 1992). In extreme instances, filter feeders may become excluded from habitats receiving high inputs of fine sediment (e.g. Armitage and Blackburn

2001). Clogging effects have also been linked to filter feeders expelling unwanted inorganic particles (e.g. Molluscs - MacIsaac and Rocha 1995), however the causal mechanism behind this response is likely associated with the relative decrease in concentration of particulate organic matter within the suspended sediment.

Abrasion caused by fine sediment has been referred to in the literature multiple times, yet the primary scientific evidence appears limited. First reported to affect macrophytes subject to excessive SSC downstream of mining activities (Lewis 1973b, 1973a), abrasion has been cited as affecting benthic assemblages and algae (Bond and Downes 2003, Francoeur and Biggs 2006) and can cause damage to soft tissues and gills in fish (Herbert and Merkins 1961, Kemp et al. 2011) and fine and fleshy body parts in macroinvertebrates (Jones et al. 2012b, Wharton, Mohajeri, and Righetti 2017). The abrasion hypothesis has been linked to behavioural responses, such as, retraction of feeding apparatus or changes to feeding mechanisms, avoidance behaviour, and passive or active drift (Bilotta and Brazier 2008).

Abrasion and clogging as primary impacts of fine sediment on macroinvertebrates remains largely hypothetical and based on correlative evidence due to the difficulties of quantifying the physical effects in real time by direct observation (Jones et al. 2012b). This study aims to build on more specific exposure experiments, such as those described by Rosewarne et al. (2014) who exposed white-clawed crayfish (*Austropotamobius pallipes*) and signal crayfish (*Pacifasticus leniusculus*) to varying concentrations of fine sediment.

# 4.2 Research aims

The aim of this chapter is to investigate the physical effects of fine sediment carried in suspension on macroinvertebrate gills of three species with varying gill morphologies (Figure 4.1); branched gills of *Hydropsyche siltalai* (Trichoptera: Hydropsychidae), feathery gills of *Ephemera danica* 

(Ephemeroptera: Ephemeridae) and plate gills (not tufted filaments) of *Ecdyonurus venosus* (Ephemeroptera: Heptagenidae). This chapter will:

- Characterise and quantify any potential damage to macroinvertebrate gills through sediment coverage of organisms or gills or abrasion of gill surfaces
- Investigate the effect of increasing suspended sediment and flow velocity on the extent of physical cover and damage observed
- Assess whether physical damage varies between gill type and structure (i.e. species).

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Figure 4.1 – Macroinvertebrate photos (left) and corresponding high-powered scanning electron microscope images (right) of the gill structure of the three test species (a) and (b) *Ecdyonurus venosus* (120 X magnification), (c) and (d) *Ephemera danica* (150 X magnification), and (e) and (f) *Hydropsyche siltalai* (200 X magnification). Black circle on (a), (c) and (e) showing location of gill(s). Figure sources (a) A. Mogliotti, EuroflyAngler.com (b) N. Phillips, UK-Wildlife.co.uk, and (c) Urmas Kruus.
It is hypothesised that physical effects would be influenced by both SSC and flow velocity. Specifically, it is predicted that coverage of fine sediment on gill surfaces would increase at higher SSC. However, the effect of increasing velocity could provide a cleaning effect. Damage associated with abrasion would be greater at higher flow velocities as a result of the higher impact velocity of sediment particles. Observing the effects of fine sediment on live macroinvertebrates presents unique challenges due to known behavioural responses to disturbance. During exposure to fine sediment in the experimental procedure, live individuals may attempt drift or seek refuge on the bed or margins of the flume (Bilotta and Brazier 2008). Alternatively, the use of microcosms to restrict movement within a defined area would have resulted in disruption of hydraulic characteristics. In both instances, live individuals would be free to move, change body position and find the most preferable refuge location within the flume in order to avoid the potential physical effects of fine sediment. As a direct result of the potential confounding effects due to the movement and avoidance behaviour (including drift out of the flume) of live invertebrates, it was decided to use immobile cadavers to provide control over the nature of exposure to elevated suspended sediment (location in the main flow, body position and alignment in relation to flow direction). This control ensured that all of the invertebrates (and hence gills) were exposed to the main flow and sediment within the flume in a similar manner throughout the experimental period, providing a benchmark from which we could determine any physical effect of fine sediment on gill surfaces.

#### 4.3 Methods

Macroinvertebrate specimens were collected from a second order lowland gravel bed stream (Woodbrook, Leicestershire, UK, 52°75' N, -1°21'W) in May – June 2017. Substrata were gently disturbed and drifting insects captured with a pond net (mesh size 1 mm) thereby minimising damage to gills. Specimens were immediately transferred to 70% industrial methylated spirit (IMS). All cadavers were examined under a dissecting microscope prior to use in experiments to ensure that gills were intact and that there was no obvious

damage to the gill structures. All individuals were late instars, given that sampling occurred in Spring, and experiments were carried out in July 2017.

Cadavers were exposed to three SSC levels (mean  $\pm$  SD): 3.5 $\pm$ 0.96 mg l<sup>-1</sup> (control), 83.7  $\pm$  7.74 mg l<sup>-1</sup> (low) and 404.0  $\pm$  77.25 mg l<sup>-1</sup> (high); and two flow velocities (0.19 m s<sup>-1</sup> and 0.37 m s<sup>-1</sup>) in a full factorial design. Due to the difficulties in measuring SSC continuously, turbidity was used as a surrogate to monitor the levels during the trials. The three SSC levels corresponded to turbidity values of <2.5 NTU (control), 100 NTU and 400 NTU (see Chapter 2 Section 2.4.1.1 for description of NTU). The SSC levels were selected to represent the range of natural conditions typically encountered in lowland UK rivers (Bilotta et al. 2012, Grove et al. 2015), and flow velocities representative of the preferences of the taxa used (Tachet et al. 2010).

In a pilot study, SEM images from individuals of *E. venosus* taken directly from the sampling location were compared with individuals which had undergone suspended sediment exposure in the flume system (at 100 NTU and low velocity treatment 0.19 m s<sup>-1</sup>). The resulting images showed that the gills from individuals examined directly from the sampling site were devoid of sediment coverage, compared to those exposed to suspended sediment in the experimental procedure which were covered with fine sediment (Appendix 2.1). This confirmed that a) the sampling location had low natural suspended sediment (at least antecedent to the sampling event), and b) the sampling method did not introduce sediment to the macroinvertebrate gills both of which could confound the results.

## 4.3.1 Experimental procedure

Experiments were conducted in a recirculating flume system at Loughborough University (flume dimensions 10 m long x 0.3 m wide x 0.5 m deep; Figure 4.2). The flume was filled with tap water and water temperature allowed to fluctuate under ambient air conditions ( $21.47 \pm 0.60$  °C). Macroinvertebrate cadavers were pinned to cork tiles (300 mm x 300 mm) fitted flush to the base of the flume. The macroinvertebrates were pinned 'facing forward' with each individual's anterior end closest to the header tank and the ventral edge in

contact with the cork tile. Each experimental trial exposed macroinvertebrate cadavers for six hours. Each experimental trial was run only once and six individuals of each species were exposed during each treatment. The experimental area (i.e. cork tiles) was located 6 m from the header tank. Textured sand boards were placed around the experimental area to create natural surface roughness and turbulence. The cadavers were located in the central third of the experimental area and each cadaver was positioned  $\sim 3.5$ times their average body length away from each other in two rows. This configuration mitigated any hydraulic effects from the flume walls and ensured fully developed flow over the experimental area (Lacey et al. 2012). Given that the configuration was based on empirical scaling's describing the dimensions of micro and macroturbulent structures around bluff bodies (Table 4.1) it also mitigated for any hydraulic effects between cadavers in the same experimental run. Given the configuration of the flume and the spacing between cadavers and solid boundaries, each cadaver can be considered statistically independent within the same trial. Following the experimental run, cadavers were carefully removed and placed in individual vials of 70% IMS. It is recognised that some sediment loss could occur during preservation (into the vial of IMS), however any sediment loss would be consistent across all trials as all individuals were preserved in the same way.



Figure 4.2 – Schematic diagram showing the experimental set up of the recirculating flume system. Macroinvertebrates were pinned across two cork tiles in the area labelled as the 'experimental area'.

Table 4.1 – Dimensions of macroturbulent structures from laboratory experiments. Dimensions scaled by flow depth (\*scaled by insect body length for current study). From Wilkes et al. (2013).

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For the SSC treatments, a fluvial sediment aggregate mixture (average organic component of 7.70±1.16%, particle size D<sub>10</sub> 10.41 µm, D<sub>50</sub> 221.40 µm, D<sub>90</sub> 505.43 µm; see below for particle size analysis method) was gradually wet sieved to 500 µm (flume limit) directly into the holding tank until the required turbidity was achieved. Turbidity was monitored at 1 s intervals using a Eureka 2 Manta sonde fitted with a self-wiping function (International Organisation for Standardisation 7027; 0-3000 NTU, quoted error ± 1%) to ensure turbidity remained consistent throughout the experimental period of six hours. If levels dropped below 95% of the target value, additional fines were added as required. The turbidity would initially peak after sediment addition and, as such, time was allowed for mixing between each new addition. Turbidity levels were stabilised at the required level before the start of each experimental trial. Despite excluding larger fractions of fine sediment (0.5 mm - 2 mm), this provided an opportunity for creating conditions analogous to natural riverine conditions since it is this finer fraction which dominates suspended sediment load (Church, Mclean, and Wolcott 1987, Chang 1998). The depth of water within the flume

was maintained at 100 mm ( $\pm$  10 mm) above the bed and velocity measured using the six-tenths-depth (Rantz 1982) at 12 locations over the experimental area (Valeport electromagnetic current meter) during each trial.

Turbidity measurements are sensitive to the physical characteristics of the sediment (Bilotta and Brazier 2008) and therefore SSC was measured for validation (Appendix 2.2). During each experimental trial, three 1 I samples of water were collected from the flume immediately downstream of the experimental area. This procedure was repeated three times for each trial (just once for the control). Samples were filtered using Whatman 0.7 µm glass microfiber filters and analysed for dry weight mass including percent organic matter through loss-on-ignition (Dean 1974). Laser particle size analysis (Malvern Mastersizer 2000) was used to obtain the particle size distribution of the sieved sediment aggregate mix (<500  $\mu$ m). The sediment was prepared by first removing organic matter by adding 5 ml of 30 % hydrogen peroxide to ~ 0.5 g sediment in a test tube. After 24 hours, the samples were heated to 70 °C until no gas bubbles were released from the mixture. Five ml of 3% sodium hexametaphosphate (Calgon) was added to disperse the particles (Gray, Pasternack, and Watson 2010). Each sample was subjected to two minutes of ultrasonic dispersion immediately prior to analysis and measured for a total of 60 s at 8-12% obscuration (Blott et al. 2004) (a particle size distribution curve is provided in Appendix 2.3).

#### 4.3.2 Microscopy procedure

For an overview of sediment coverage on macroinvertebrate gill surfaces, individual gills from cadavers within each treatment were mounted on microscope slides using Hoyer's solution. Images of the gills from each slide were examined under a stage microscope. Images were taken using a Nikon eclipse 80i (for examples see Appendix 2.4). The fine sediment accumulation on each individual gill was visually assessed qualitatively by examining individuals used in experiments using a dissecting microscope and found to be consistent across all gills of each individual, within each treatment. Due to this consistency, only two gills from a single individual of each species from each treatment were used for detailed examination.

For detailed gill surface profile images, Scanning Electron Microscopy (SEM) was used. Individual gills were removed from cadavers from each experimental trial and prepared by freeze-drying overnight (CHRIST BETA 1-8 LDplus Freeze Drier). For *E. venosus*, gills five and six were used, whereas gills five and eight were used for *H. siltalai* and gills four and six for *E. danica*. The selection of these particular gills was made because they were intact across all individuals within each species. Gill loss was not considered as an impact within the present study as the selected taxa have more robust gills compared to other taxa which commonly lose tracheal gills during disturbance (e.g. Baetidae and Leptophlebidae; Elliott and Humpesch 2010). An additional step was required to prepare gills for the investigation of physical damage by abrasion, in order to remove the fine sediment adhered to the surface of the gills. One individual of each species from each treatment was placed in an ultrasonic bath (Fisherbrand FB11004) for two 30 s periods on setting five. Gills were sputter-coated in Gold-Palladium for 90 s prior to analysis.

Images were captured on areas of the gill surface where the following criteria were satisfied: the gill surface filled the whole frame; the aspect of the surface was normal to the optical axis; and the area was representative of the coverage on the gill surface and away from the gill margin. To meet this criteria, three images were taken of each gill, at different locations on the surface, at 5,000 X magnification for *E. venosus* and *E. danica* and the higher magnification of 25,000 X for the smaller gills of *H. siltalai*. However, some SEM images did not meet these criteria and were discarded. For images used to quantify sediment coverage of gill surfaces, this left 31 images for *E. danica*, 33 for *E. venosus* and 36 for *H. siltalai*. All images were retained for assessing physical damage by abrasion because the criteria were less relevant for qualitative observations of abrasion (36 for each species).

In order to determine the appearance of sediment particles, fine sediment samples collected from the macroinvertebrate sample site in the field (during

macroinvertebrate collection) and from the experimental sediment aggregate mix were oven-dried overnight, sieved to 500 µm and processed for SEM examination using the method outlined above.

## 4.3.3 Image analysis

The resulting images were used to characterise the extent of surface coverage and abrasion. To reduce subjectivity from visual estimates, a non-automated digital image analysis technique developed and described in Turley et al. (2017) was used. The method was developed in order to reduce variability from estimate-based methods and has been shown to have low inter-operator variability. Areas of sediment coverage were highlighted by the same operator throughout the process using the foreground colour (#FA0200) in Adobe Photoshop (v19.0) (Figure 4.3). Each image was then exported and uploaded to *PixelCount* (Turley, M. D. et al. 2017), a software application that calculates the percentage of each image highlighted in a selected colour, thereby providing the percentage of sediment cover on each image. Bacteria on the gill surfaces, identified as rod-shaped particles (Lemly 1982), were not highlighted. Bacteria are likely to be a part of the natural flora of live individuals (Lemly 1998, 2000, Lemly and King 2000), and not part of bacterial activity which occurred during degradation. Abrasion was assessed using a qualitative visual estimate of the images, areas of abnormal gill surface textures and marks were recorded.

#### 4.3.4 Statistical analysis

A three-way unbalanced ANOVA (Akritas, Arnold, and Brunner 1997) was used to test for significant effects of species, SSC, flow velocity and all interactions in relation to the surface area of the gill image covered by fine sediment. Percentage data (percentage of sediment coverage) was arcsine square root transformed prior to analysis as the appropriate transformation for proportional data to meet the assumptions of the ANOVA test (Sokal and Rohlf 1995). The resulting nested models were compared separately for each species using an F-test. Pairwise post-hoc Tukey's tests were carried out using the *glhtfunction* from the *multcomp* package (Hothorn, Bretz, and Westfall 2008). Given the relatively small sample size, and the fact that fine sediment accumulation was consistent across all gills of each individual within each treatment, gill number was not included as a random effect. All statistical analysis was carried out using R version 3.5.3 (R Development Core Team 2019).



Ecdyonurus venosus 8.91%

Ephemera danica 18.85%

Hydropsyche siltalai 49.59%

Figure 4.3 – Scanning Electron Microscopy images of each test species; *Ecdyonurus venosus* (5,000 X magnification), *Ephemera danica* (5000 X magnification) and *Hydropscyhe siltalai* (25,000 X magnification). Original scanning electron microscopy images (top row) and the same images after digital image analysis (with sediment particles highlighted in red) (bottom row). The percentages equate to the total area per image covered with fine sediment.

# 4.4 Results

The physical effects of fine sediment on the individual gill tissues predominantly consisted of coverage of fine sediment on the gill surface (Figure 4.4). Numerous chloride cells were observed on the SEM images of both *E. danica* and *E. venosus* (white circles, Figure 4.4). For *E. danica* these were covered by sediment for all concentrations, but for *E. venosus* these remained clear for the control conditions. The texture of sediment particles covering gills was consistent with that of the fine sediment particles collected from the macroinvertebrate sample sites and from the experimental sediment aggregate mix (Figure 4.5).



Figure 4.4 – Examples of Scanning Electron Microscope images for *Ecdyonurus venosus* (5,000 X magnification), *Ephemera danica* (5000 X magnification) and *Hydropscyhe siltalai* (25,000 X magnification) after exposure to two controls and four treatments of varying SSC and flow velocity. Control (1) = 3.5 mg l<sup>-1</sup> SSC at 0.19 m s<sup>-1</sup>, control (2) = 3.5 mg l<sup>-1</sup> SSC at 0.37 m s<sup>-1</sup>, treatment (3) = 83.7 mg l<sup>-1</sup> SSC at 0.19 m s<sup>-1</sup>, treatment (4) = 83.7 mg l<sup>-1</sup> SSC at 0.37 m s<sup>-1</sup>, treatment (5) = 404.0 mg l<sup>-1</sup> SSC at 0.19 m s<sup>-1</sup> and treatment (6) = 404.0 mg l<sup>-1</sup> SSC at 0.37 m s<sup>-1</sup>. An example of a chloride cell is circled in white for the two Ephemeroptera species, *E. venosus* and *E. danica,* in the images from treatment one.



Figure 4.5 - Scanning Electron Microscope images of the sediment aggregate mix (used in the experimental treatments – top) and natural riverine sediment (collected from the macroinvertebrate collection sites – bottom) at increasing magnifications (left to right); 100 X, 5,000 X and 10,000 X.

The extent to which the gill was covered varied by SSC and species (Figure 4.6). A three-way ANOVA demonstrated sediment cover on the gill surface did significantly vary as a function of species (F = 29.50, p < 0.001), sediment (F =21.41, p <0.001), and species:sediment (F = 8.67, p <0.001), species:velocity (F = 5.67, p < 0.001) and three-way (F = 5.62, p < 0.001) interactions (Appendix 2.5). The sediment:velocity interaction was not significant (F=0.96, p=0.39) across all species. Neither was this interaction significant for *E. venosus* (F = 1.53, p = 0.23) or *E. danica* (F = 1.37, p = 0.27). However, the model including the sediment:velocity interaction for *H. siltalai* was significant (F = 9.76, p <0.001) (Appendix 2.6). Post-hoc tests indicated significantly more fine sediment coverage for E. venosus as SSC levels increased but no significant effect of velocity (Table 4.2). In contrast, there were no significant effects of either SSC or flow velocity on gill cover on E. danica. The only significant result for *H. siltalai* was a significant increase in fine sediment coverage between low (83.7 mg l<sup>-1</sup>) and high SSC (404.0 mg l<sup>-1</sup>) only when velocity was low (0.19 m s<sup>-1</sup>) <sup>1</sup>) (Figure 4.6; Table 4.2).

Physical damage in the form of abrasion was evident in two images, one for *E. venosus* and one for *E. danica*. In these instances, marks on the surface of gills appeared to be inconsistent with normal gill texture appearance, potentially

indicating abrasion from sediment particles (Figure 4.7). No abrasion was observed on gills of *H. siltalai*.



Suspended sediment concentration

Figure 4.6 – Boxplots of percentage gill coverage between experimental trials for *Ecdyonurus venosus* (a), *Ephemera danica* (b) *Hydropscyhe siltalai* (c).

Table 4.2 - Summary results from the post-hoc general linear hypothesis tests to determine the effects of sediment and velocity treatments on percentage gill coverage for each of the three test species. \*Denotes a significant term (p <0.05).

Hypothesis	Estimate	SE	t	р
			value	
E. venosus				
Sediment: 404.0 mg $I^{-1}$ – Control = 0	0.530	0.053	9.975	<0.001*
Sediment: 83.7 mg $I^{-1}$ – Control = 0	0.308	0.054	5.663	<0.001*
Sediment: 83.7 mg $l^{-1}$ – 404.0 mg $l^{-1}$ = 0	-0.222	0.052	-4.296	<0.001*
Velocity: 0.19 m s <sup>-1</sup> – 0.37 m s <sup>-1</sup> = 0	-0.095	0.043	-2.193	0.121
E. danica				
Sediment: 404.0 mg $I^{-1}$ – Control = 0	0.019	0.087	0.217	0.995
Sediment: 83.7 mg $l^{-1}$ – Control = 0	-0.087	0.088	-0.983	0.724
Sediment: 83.7 mg $l^{-1} - 404.0$ mg $l^{-1} = 0$	-0.106	0.079	-1.331	0.503
Velocity: 0.19 m s <sup>-1</sup> – 0.37 m s <sup>-1</sup> = 0	0.153	0.068	2.233	0.113
H. siltalai				
$0.19 \text{ m s}^{-1}$ : 404.0 mg l <sup>-1</sup> – Control = 0	0.216	0.087	2.491	0.091
$0.19 \text{ m s}^{-1}$ : 83.7 mg l <sup>-1</sup> – Control = 0	-0.216	0.087	-2.501	0.089
$0.19 \text{ m s}^{-1}$ : 83.7 mg l <sup>-1</sup> – 404.0 mg l <sup>-1</sup> =	-0.433	0.087	-4.992	<0.001*
0				
$0.37 \text{ m s}^{-1}$ : 404.0 mg l <sup>-1</sup> – Control = 0	-0.029	0.087	-0.343	0.996
$0.37 \text{ m s}^{-1}$ : 83.7 mg l <sup>-1</sup> – Control = 0	0.078	0.087	0.904	0.867
$0.37 \text{ m s}^{-1}$ : 83.7 mg l <sup>-1</sup> – 404.0 mg l <sup>-1</sup> =	0.108	0.087	1.247	0.673



Figure 4.7 - Possible evidence of abrasion seen as striations (within white circled areas) on a) *Ephemera danica* (83.7 mg  $I^{-1}$  SSC and 0.19 m s<sup>-1</sup> without ultrasonic treatment, image at 5000 X) and b) *Ecdyonurus venosus* (3.5 mg  $I^{-1}$  SSC and 0.37 m s<sup>-1</sup> with ultrasonic treatment, image at 10,000 X).

## 4.5 Discussion

This study aimed to investigate the physical effects of suspended fine sediment at differing flow velocities on the gills of three common species of lotic macroinvertebrates. It was hypothesised that increasing SSC and flow velocity would affect the extent of physical damage in the form of sediment coverage of macroinvertebrate gill surfaces. Evidence was found that partially supports this, with gill coverage in *E. venosus* increasing significantly with SSC. Gill coverage in *H. siltalai* was only significantly different between low and high SSC treatments when flow velocity was low. There was no effect of any SSC on gill coverage in *E. danica*. It was also hypothesised that increasing SSC and velocity would lead to increased abrasive damage to gill surfaces. Potential abrasion was only observed in two instances. The striations observed in these instances could be a result of gill abnormalities or damage caused by other mechanisms. Hence, there is little support for this second hypothesis.

Fine sediment coverage in *E. venosus* appeared to increase proportionately with SSC. The gills of *E. danica* were consistently covered with fine sediment across all three SSC treatments. The fine sediment coverage of *H. siltalai* gills appeared linear when the flow velocity was slower. However, this relationship

was not observed at the higher flow velocity. Species identity was significant in predicting sediment cover, and gills of *H. siltalai* had lower sediment coverage across all the treatments compared to the other species.

In the closed tracheal system of aquatic insects, respiration occurs through tracheal gills which vary in structure by macroinvertebrate order and family level. This variation can partially help explain the results recorded. All six pairs of *E. danica* gills are bilamellated, feather-like and oscillate in synchronous pairs, creating a water current over the dorsal side of the body between the two rows of gills (Eastham 1939). During the experimental procedure, gills were positioned upwards perpendicular to the body in the water column, directly exposed to fine sediment in suspension and saltating over the bottom of the flume. The small feathering branches on each tracheate gill effectively became nets for fine sediment, with high sediment coverage recorded even for the control trials. Ecdyonurus venosus gills are held to the side of the abdomen and project downwards. Pairs 1-6 consist of a lamelliform gill plate and a proximal gill tuft underneath, whilst gill 7 comprises a single gill plate (Eastham 1937). The gill plate was analysed for the study as this portion of the tracheal gill is exposed to the flow and fine sediment in suspension. Visual observations showed the gills stayed relatively stationary during the experimental procedure, in contrast to *E. danica*, and exhibited increasing sediment coverage with SSC. *H.siltalai* gills consist of a few, pale, branched gill tufts held under the abdomen. This species exhibited lower gill sediment coverage than the two Ephemeroptera species. The mechanism for this is likely due to the gill location under the abdomen which allows the body to protect the gill from fine sediment.

#### 4.5.1 Ecological interpretations

It should be noted that for the practicalities of this study, cadavers were used to determine the physical effects of suspended sediment on macroinvertebrates (gill coverage and abrasion). Where historically the deposition of particles on the surface of gills has been defined as 'clogging', the potential damage in this study was defined as fine sediment 'coverage' of gills. This is because it cannot be confirmed whether sediment coverage on gill surfaces directly equates to

impaired functioning of key structures involved in respiration and osmoregulation through the use of cadavers. Additionally, the individuals were not able to exhibit avoidance behaviours such as active drift (Doeg and Milledge 1991, Larsen and Ormerod 2010) or clean sediment covered structures (Eastham 1939). However, the results from this study are intuitive based on the traits and preferences of the test species which is explained below (Figure 4.8).

*Ephemera danica* gills were covered with fine sediment consistently, regardless of the experimental trial. This species displays a habitat preference for sand, silt and clay substrates within which the organism burrows (Elliott and Humpesch 2010). All *Ephemera spp.* display trait characteristics associated with life in fine sediment deposits, with modified mouthparts, processes (appendages) on the head, and broadened prothoracic legs which allow them to excavate and burrow into the substrate (Eriksen 1963, Elliott and Humpesch 2010). The presence of numerous hairs on the gills prevent fine sediment particles from completely smothering them (Hynes 1970) and the setae brushes on the rear legs are used to clear body parts of accumulated debris (Eastham 1939). Based on these behaviours and traits, *Ephemera danica* is therefore considered relatively tolerant of excessive fine sediment (Bennett 2007, Extence et al. 2013). As coverage of sediment on *E. danica* was consistent across all treatments, it is possible that they are not adapted to avoid accumulation and can rather tolerate accumulation to an extent possibly in part by cleaning gills using their rear legs.

*Ecdyonurus venosus* is widely described as a clinger and lives on rocks and other hard substrates. It is adapted to live in close association with high flow velocities and shear stresses (Lancaster and Belyea 2006), and avoids dislodgment from substrates by being dorsoventrally flattened and possessing large curved tarsal claws to cling on to hard substrates (Wichard, Arens, and Eisenbeis 2002, Elliott and Humpesch 2010). The role of its lamelliform gill is to generate a current and draw oxygen in, whereas the filamentous sections are for respiration (Eastham 1937). For *E. venosus*, the lamelliform gill provides some protection from fine sediment to the filamentous gills underneath. Based on their body plan and behaviour, the gills are relatively protected at low sediment concentrations (compared to *E. danica* gills which project upwards

into the water column), however they are susceptible to increasing fine sediment and potentially a mechanism to remove fine sediment accumulation. Consistent with these characteristics and the results of previous biomonitoring studies (e.g. Murphy et al. 2015; Turley et al. 2016), the findings support the classification of *E. venosus* as sensitive to fine sediment (Figure 4.8a).



Figure 4.8 - Ecological information for the three test species (*Ecdyonurus* given at genus level due to lack of availability of data at species level). Sensitivity of each test species for biomonitoring indices WHPT, EPSI and CoFSI (see Chapter 2 Section 2.4.2 for index descriptions) (a) and trait preferences (Tachet et al. 2010) for habitat (b), velocity (c) and mode of locomotion and relationship to substrate (d). *Ecdyonurus* and *E. danica* have identical trait preferences for mode of locomotion and relationship to substrate and therefore overlap in (d).

*Hydropsyche siltalai* typically constructs feeding nets either side of a tubular retreat (Edington and Hildrew 1995). These structures are either exposed (at right angles to the local flow) or in crevices beneath and underneath stones where gravel and plant material can be used as support. Particles caught in the net are collected using the mandibles and prothoracic legs, whilst inedible particles are ejected (Edington and Hildrew 1995). In environments characterised by high availability of fine sediment, these nets become clogged causing the organism to spend increasing amounts of time cleaning the nets or in extreme instances abandoning the nets (Strand and Merritt 1997). Although it is regarded as moderately sensitive to fine sediment (Murphy et al. 2015, Turley et al. 2016), *H. siltalai* had relatively low coverage of sediment of gills across all trials, suggesting that sensitivity in this species is probably primarily associated with the filter feeding mechanism and/or cleaning of nets.

#### 4.5.2 Potential biological implications

Due to the use of cadavers in this experiment, biological impacts remain uncertain. However, some speculation can be made based on the roles of macroinvertebrate gills and the importance of proper functioning. Respiration and osmoregulation are intimately associated processes in aquatic organisms and essential to inhabiting aquatic environments (Wichard, Arens, and Eisenbeis 2002). Osmoregulation is required to balance the concentration of ions and fluids in an insect's body. During respiration, through the diffusion of oxygen into the insect, water also penetrates by osmosis. Excess water is excreted by the body and the re-uptake of ions is carried out by specialised chloride cells (Figure 4.9) which are usually located on the gills.

Chloride cells which become clogged with fine sediment will ultimately affect osmoregulation (Bruton 1985, Waters 1995, Bergstedt and Bergersen 1997). However, chloride cells can vary in number depending on water salinity in order to continue to balance osmoregulation in sub-optimal conditions (Wichard, Tsui, and Komnick 1973). It could therefore be possible that at continually high SSC levels when gills are likely to be heavily covered by fine sediment (and function inhibited), chloride cell densities can increase so osmoregulation can continue

to be carried out effectively. Trichopterans do not possess chloride cells and instead the uptake of ions is carried out by other forms, predominantly through chloride epithelia (Wichard, Tsui, and Komnick 1973, Wichard, Arens, and Eisenbeis 2002). Possessing a range of methods of ion re-uptake may indicate osmoregulation is less affected by fine sediment deposition and coverage of gills and other body parts for trichopterans. Whilst studying the effect of aluminium on gills of *E. danica*, Herrmann and Andersson (1986) noted mucus formation on the gills during exposure. The result of this mucus formation was to impair osmoregulation and lower respiration efficiency, causing the mayfly to increase respiration to compensate. It is unknown whether insect larvae can secrete mucus for gill protection as a result of abrading sediment, as is the case for fish gills (McCubbin et al. 1990). Although, in high sediment conditions, the mucus secretion can result in increased susceptibility to coverage of sediment on the gill surface and ultimately suffocation of the fish.

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Figure 4.9 – Illustration of chloride cells in mayfly larvae: a) single cells and b) cell complexes (from Wichard and Komnick 1973; Wichard, Arens and Eisenbeis 2002).

# 4.6 Conclusion

This study provides evidence of the effect of varying levels of fine sediment in suspension on macroinvertebrate gills of specific taxa using a novel

methodological approach, through SEM and image analysis, that can be applied in freshwater research to produce quantifiable results. It is recognised that there is some subjectivity in the imaging process, although the systematic digital image analysis method employed minimises such subjectivity in the assessment of fine sediment coverage. It is therefore suggested that this SEM application provides a robust estimate of fine sediment coverage of gill surfaces. The results should be interpreted with care when applied to natural conditions due to the experimental use of cadavers. In order to develop this research further, developing a way in which live specimens could be used in a flume set up to more accurately replicate natural conditions is recommended. Overcoming this limitation could potentially enable field experiments in mesocosm systems where the effects of sediment coverage across natural sediment flux dynamics (i.e. rising limb, peak, and falling limb) could be determined. Closed chamber respiration methods, using live insects, could be used to confirm whether fine sediment coverage on insect gills has a negative effect on respiration (Rostgaard and Jacobsen 2005). Abrasion appeared to be less important when considering the effects of physical damage from fine sediment, although further research is required to assess its prevalence with varying levels of angularity, particle size and water velocities. Additionally, the interaction of sediment particle size relative to macroinvertebrate body size could have implications to the extent of physical damage and associated negative effects. This research will help us understand how invertebrates respond to excess fine sediment and the traits that should be considered to improve fine sediment-specific biomonitoring tools (Wilkes et al. 2017). For example, 'gill type' could be incorporated as a new trait group.

Studies assessing the direct and physical impacts of fine sediment for macroinvertebrates at the organism level have been relatively limited to date. This experiment has, for the first time, demonstrated the potential physical effects of fine sediment on macroinvertebrate gill tissue, through fine sediment coverage and abrasion, in three species of lotic macroinvertebrates. In contrast to the widely cited effects of abrasion in the literature, the results showed evidence that gill coverage was the primary effect. However, the increasing

SSC was associated with increased gill coverage for only one species (*E. venosus*). Flow velocity and species' traits and ecology interacted to produce a variable response to fine sediment. Although these results must be observed with caution given the use of cadavers, these differences can be explained by variations in gill structure, and in relation to known species' habitat preferences and traits.

# Chapter 5 – Testing the performance of biotic indices for fine sediment-specific biomonitoring

## **Chapter overview**

Appropriate monitoring practices are required in order to efficiently identify ecologically impacted water bodies. In Chapter 2, the effects of fine sediment on macroinvertebrates, and the importance of macroinvertebrates as agencies for biomonitoring, were discussed. This chapter begins by describing, in detail, the development of sediment-specific biomonitoring indices in the UK, whilst also highlighting their key methodological differences. Through a national fieldwork sampling regime, these existing sediment-specific biomonitoring indices were tested along with indices for general ecological health. Trait-based approaches were incorporated into the analysis in line with emerging evidence on the guantitative use of traits in the development of biomonitoring indices. Further insights into the response of macroinvertebrates to fine sediment metrics were explored using a variety of statistical techniques such as threshold indicator taxa analysis, fourth corner analysis and gradient forest. The results show reach scale visual estimates of fine sediment to be a suitable proxy for timeconsuming fully quantitative estimates of total surface sediment. PSI derived sediment-specific biomonitoring indices (PSI, EPSI and in particular the EPSImixed) showed a closer association with different metrics of fine sediment than CoFSI. However, the majority of variation in sediment-specific index scores at each site were related to habitat and flow variables. Gradient forest analysis showed shredders to be the most sensitive trait modality to fine sediment, supporting conclusions made in Chapter 3. The combined results of this chapter point to the accomplishment of sediment-specific indices that can detect sediment stress, a pressure response which is closely synonymous with organic stress, and in a community predominantly controlled by flow.

# 5.1 Introduction

# 5.1.1 National developments in fine sediment-specific biomonitoring tools

Chapter 2 (Section 2.4.2) introduced the development of several sedimentspecific biomonitoring indices and described some examples of those used globally. The sediment-specific biotic indices developed for use in the UK are: The Proportion of Sediment-sensitive Invertebrates group (PSI; Extence et al. 2013), its empirical development (EPSI; Turley et al. 2015, 2016), and the Combined Fine Sediment Index (CoFSI; Murphy et al. 2015). Each index was developed using contrasting methods making direct comparison difficult (Table 5.1). However, it is important to dissect these differences to understand how these indices work and how they compare in terms of performance.

Index	Fine sediment metric	Method of development	Number of scoring taxa	
PSI	Percentage fine grained	Expert knowledge	1030	
(Extence et	sediment (visual			
al. 2013)	estimates)			
EPSI	Percentage fine grained	Expert knowledge	433	
(Turley et al.	sediment (visual	and empirical		
2015)	estimates)	weightings		
CoFSI	Total fine-grained	Entirely empirical	105	
(Murphy et	sediment mass			
al. 2015)	(resuspension method)			
EPSImixed	Percentage fine grained	Expert knowledge	355	
(Turley et al.	sediment (visual	and empirical		
2016)	estimates)	weightings		

Table 5.1 – A summary of the four main indices (in chronological order) developed for use in sediment specific biomonitoring for use in England.

The first sediment sensitive index developed for application in the UK, PSI (Extence et al. 2013), used expert opinion to assign individual taxa to one of four Fine Sediment Sensitivity Ratings (FSSR). The assignment of FSSR was

based on an extensive literature review and the assessment of the functional traits of each individual taxon. To calculate the PSI score for a specific site, the FSSR scores are weighted based on the abundance of scoring taxa sampled using a standard kick net procedure. The PSI score is calculated as a percentage based on the scores for sediment sensitive groups over the scores of all groups (see Appendix 3.1, Table A3.1 for sensitivity categories and abundance weightings, and Equation A5.1 for index formula). This method has a sound biological basis and has been used for the development of many other well-established biotic indices (e.g. the Lotic Index for Flow Evaluation index; Extence, Balbi, and Chadd 1999). However, there are known difficulties of using expert judgement to develop biotic indices, one of which is allocation error of sensitivity level (Walley and Hawkes 1996).

The EPSI index (Turley et al. 2015) is based on the PSI index but is enhanced by adding an empirical weighting element to increase its predictive power (see Appendix 3.1 Equation 3.2). In the original PSI index, all taxa within the same FSSR were considered to be equally tolerant or sensitive to fine sediment. The development of EPSI involved fitting empirical weightings based on large-scale field observations (Turley et al. 2015, 2016). Adding this empirical element to an index that was purely theoretical arguably makes the index more robust. However, the individual species scores were constrained by the original FSSR allocated to the species in the PSI index development. The empirical weightings were derived from non-linear optimisation methods using the RIVPACS IV dataset (NERC [CEH] 2006). In a previous publication the same authors found visual estimates (of fine sediment on the stream bed within the sampling area) to be most related to PSI scores (Turley et al. 2014) and therefore visual estimates of fine sediment (sand, silt and clay <2 mm diameter) were used in the empirical optimisation process. Visual estimates of fine sediment are a semi-quantitative assessment and have been found to have high inter-user variability (Murphy et al. 2015) and can be highly influenced by depth, light penetration and turbidity. Additionally, the visual estimate method only assesses the surface drape of fine sediment which can be unrelated to the ingress of fines (Murphy et al. 2015). The authors do recognise this limitation and cite

Glendell et al. (2014), stating that the method '*does provide a measure of the percentage cover, which theoretically, should be related to the PSI index*' (Turley et al. 2014, p. 2270). Nonetheless, this is an assumption that has not been tested. Such potential weaknesses in methodology could lead to bias in the measurement of total fine sediment at each site. In turn, this could result in poor associations between fine sediment and macroinvertebrate assemblages during development of the revised weightings. In order for EPSI to be better integrated into statutory monitoring, a more recent iteration of the index containing mixed level taxon scoring has been published (EPSImixed; Turley et al. 2016).

In contrast to the PSI and EPSI indices, the development of CoFSI (Murphy et al. 2015) was entirely empirical. The approach combined macroinvertebrate and sediment data collected from 179 stream sites across England and Wales. Fine sediment was collected using the resuspension method providing a quantitative assessment of fine sediment (see Section 2.4.1.2). Murphy et al. (2015) used partial canonical correspondence analysis (pCCA) to factor out the variation from normal biological variance. The analysis showed that the mass of organic sediment in erosional areas, the total mass of fine sediment in surface drapes of depositional areas and the percentage of total sediment that was organic in erosional areas explained most of the variation in macroinvertebrate assemblages. However, the eigenvalues (contribution of each variable to the explanatory power of the overall pCCA model) for each of these values were relatively low (Appendix 3.1, Table A3.2). The score given to each species was related to the position on the two axes of the pCCA model (Appendix 3.1, Equation A3.3). The respective axis, or indices, represent the organic fine sediment index (oFSI) and the total fine sediment index (ToFSI) which collectively make up CoFSI. This robust statistical approach helps to factor out confounding and collinear variation within the data. Conversely, the index does not take into account species abundance and the final calculation is based on presence/absence only. In the development of the CoFSI, organisms that occurred in fewer than 10% of all samples were excluded, eliminating 208 of 313 taxa. This potentially eliminates a large number of taxa conforming to K-

selecting life strategies (e.g. large body size, longer life expectancies, produce fewer offspring).

A key part of the development of any biomonitoring index, and even more so for pressure-specific indices, is testing against known gradients of the particular stressor to understand its predictive power, usually by correlation. Despite the different way in which CoFSI and EPSI were developed, they have remarkably similar correlations with metrics of fine sediment when tested (Table 5.2). Although stronger than PSI, the most theoretical of indices, these values are still relatively moderate despite being within the range of values for other indices used in the implementation of the Water Framework Directive (Birk et al. 2012). However, the indices are not consistently tested in the same way, i.e. against the same metric of fine sediment. It is therefore difficult to reach a conclusion over which index is performing best. This highlights the need for an independent assessment to determine how these indices respond to alternative metrics of fine sediment.

In summary, EPSI shows the strongest relationship with metrics of fine sediment. However, these values are acquired using visual estimates which is a relatively weak method of quantifying fine sediment. Additionally, visual estimates were the metric of fine sediment used in the development of the index and therefore EPSI would always be expected to exhibit a stronger relationship with this metric. Both EPSI and CoFSI have been shown to have strong correlations with LIFE (Lotic Index for Flow Evaluation; Extence, Balbi, and Chadd 1999) and WHPT\_ASPT (Walley Hawkes Paisley Trigg Average Score Per Taxon; Walley and Hawkes 1996) scores when analysed over large scales at many sites (Turley et al. 2015, 2016, Murphy et al. 2015). This is likely because taxa that are sensitive to excess fine sediment are also sensitive to the gradients measured by WHPT\_ASPT and LIFE. Flow is intrinsically linked with fine sediment dynamics in rivers. Additionally, slower flowing environments typically have less dissolved oxygen in the water column (and will be more susceptible to sediment deposition through transport limitation). The optimal index will be able to detect a particular pressure across its entire gradient regardless of comparable responses to other pressures. A full review of the two

indices (CoFSI and PSI group) incorporating a variety of fine sediment metrics is required to test their performance and understand what situations they are most appropriate for. This highlights a need to independently test these existing indices in a 'like-for-like' setting i.e. across the same sites and using the same methods of quantifying fine sediment.

Table 5.2 – Published results of Spearman's rank correlation values of sediment-specific biotic indices PSI, EPSI and CoFSI with fine sediment. This item has been removed due to third party copyright. The unabridged version of the thesis can be viewed at the Lanchester library, Coventry University

## 5.1.2 Development of trait-based approaches

Quantifying functional trait diversity within a river system may help to explain interactions within the system that have been missed using taxonomic diversity alone. This approach is possible because immediately after stress exposure, traits that impart resistance will be favoured by the community, whereas species with resilient traits will recover most rapidly following disturbance. This filtering of traits allows biomonitoring approaches *to 'yield mechanistic understanding rather than our current ability to simply observe that ecological change has occurred*' (Culp et al. 2011, p. 187). This is because detecting a functional response can provide an indication of why a change in abundance may be occurring. Doretto et al. (2018) observed that functional traits seemed to better detect impacts of stressors than traditional community indices. Therefore, indices which are based on functional traits can have a greater mechanistic basis (Wilkes et al. 2017).

The PSI index was developed based on expert knowledge of taxonomic traits. It can therefore be assumed that the PSI index, including the empirically derived EPSI, will be more mechanistic. Wilkes et al. (2017) quantitatively related fuzzy coded trait data (Tachet et al. 2010) to biotic index scores and found that the well-established, empirically developed, WHPT index had a high R<sup>2</sup> value (0.92), indicating good agreement between species traits and their scores under WHPT. Both EPSI and CoFSI showed a relatively poor fit when modelled against traits compared to WHPT, indicating lower optimisation and lack of a mechanistic basis. When considering the scores or weightings of individual taxa under both indices, there are clear discrepancies (Figure 5.1), indicating differences in the mechanistic basis of the indices. This points to a requirement for further development of trait-based biomonitoring.

Integrating macroinvertebrate traits into biomonitoring methods is an emerging practice (Doledec, Statzner, and Bournard 1999, Archaimbault et al. 2009, Doretto et al. 2018). Chapter 3 details existing evidence of trait responses to fine sediment. The results are equivocal, in part because of the lack of studies that contain this information, with the exception of shredders for which evidence points to a significant negative response to fine sediment. There has been some progress integrating traits into sediment-specific biomonitoring indices. Murphy et al. (2017) showed a potential to integrate traits in to fine sediment impacts using significant taxa-trait-environment relationships from RLQ analysis. Most recently, Doretto et al. (2018) attempted to integrate traits, along with some traditional taxonomic indices, into a multi-level metric (MMI) to assess excess sediment. The MMI included a combination of taxonomic indices (total taxa

richness and richness in Ephemeroptera, Plecoptera and Trichoptera taxa) and functional metrics (abundance of rheophilous taxa preferring coarse substrata).

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Figure 5.1 - Comparison of taxon scores under CoFSI and taxon weightings under EPSI for the 71 taxa that score under both indices (from Wilkes et al. 2017).

# 5. 2 Research aims

The current intention of UK monitoring agencies is to integrate sediment sensitive indices in to the next round of WFD (2021-2027) (Environment Agency, *pers comm*). The aspiration of the UK Technical Advisory Group (UKTAG) is to '*develop biological methods that describe the impact of sediment in fresh waters effectively*' (UKTAG 2018). EPSI is considered to have the most potential at present (John Murray-Bligh, Environment Agency Lead for Invertebrates on UKTAG Freshwater Task Team, *pers. comm*). However, there is a requirement to independently test a range of indices to understand how sediment-specific biomonitoring tools can be used to enhance monitoring and management of fine sediment.

The aims of this chapter are:

- To compare methods of measuring fine sediment (visual versus quantitative)
- Test the performance of sediment-specific biomonitoring indices and compare to the performance of other, non-sediment-specific (non-specific) indices
- Determine which environmental variables and antecedent conditions affect index performance
- Provide new insights into the mechanistic basis for fine sediment biomonitoring

These aims were achieved by conducting a new field sampling regime to collect macroinvertebrates, environmental variables (including hydrological metrics) and different metrics of fine sediment. Hypotheses (Table 5.3) were identified based on existing knowledge of sediment-specific indices and trait-sediment relationships (Section 2.3 and 3.4.2). This chapter will ultimately provide an overview of the performance of current nationally developed fine sediment biomonitoring indices and make recommendations for their use and improvement.

Table 5.3 – Table of hypotheses.

	Hypotheses
1	EPSI and CoFSI will show a stronger relationship than PSI (as EPSI and CoFSI include empirical calibrations) with metrics of fine sediment
2	EPSI will show a stronger relationship with visual estimates of fine sediment and, conversely, CoFSI will show a stronger relationship with quantitative estimates of fine sediment
3	All fine sediment-specific biomonitoring indices will show a stronger relationship with fine sediment metrics than non-specific indices
4	Shredders will be sensitive to fine sediment
5	Burrowers will be tolerant of fine sediment

# 5.3 Methods

## 5.3.1 Site selection

In order to collect data that were robust and representative of (semi-) natural conditions, the sites sampled were required to meet a pre-determined set of criteria:

- Provide a good spatial distribution across England
- Cover a range of river types characteristic of lowland UK
- Consist of a range of fine sediment pressures (e.g. high or low fine sediment pressure, high or low organic content in fines)
- Be accessible for data collection, i.e. wadeable
- Be minimally affected by disturbance from other factors which may confound the effects of fine sediment (e.g. habitat and water quality)

In order to meet the criteria, sites were selected by filtering from existing national biological monitoring locations. A data set was acquired (Lathouri and Klaar 2016) which matches Environment Agency (EA) biological and chemical monitoring sites with River Habitat Survey sites. This data set was then subjected to a filtering process (Figure 5.2).

# Phase 1 – Water quality

The list of national sites had already been screened using EA water chemistry monitoring data (WIMS) '*from 2012-2014, where a minimum of eight samples over three years was required for site classification*' (Lathouri and Klaar 2016, p5). Sites which were failing physico-chemistry status for dissolved oxygen (DO) and ammonia for one or more seasons were removed from the data set. These physico-chemical parameters were chosen based on data availability. It is recognised that these sites could have high inputs of nutrients, such as phosphorous. High nutrient inputs predominantly impact algae and plant communities, increasing growth and affecting species composition in the channel (WFD-UKTAG 2013). This can lead to wider effects on the community. However, any successive impacts, such as eutrophication leading to high

biological oxygen demands (BOD), will likely affect the overall DO concentration which has been included in the filtering process.



Figure 5.2 – Conceptual model showing the two elements which were combined to select final sites list which involved combining national data sets with a screening process (a) and detail of each of the four filtering processes which together encompass the screening process (b) see text for explanation of each screening phase).

# Phase 2 – River typology

The next step was to consider the typology of the rivers to be included in the study. In the UK, most lowland rivers are transport-limited in relation to fine sediment (Naden et al. 2016). Relatively stable seasonal flow regimes and groundwater abstraction reducing river discharges, coupled with an increase in arable farming in lowland areas, results in lowland gravel rivers being most at risk of fine sediment accumulation (Collins, Walling, and Leeks 2005). Therefore, for this study, lowland rivers were chosen as the focus. The River Invertebrate Prediction and Classification System (RIVPACS) (Wright, Furse,

and Moss 1998) uses TWINSPAN (Two Way INdicator SPecies ANalysis) to classify rivers into one of 43 end groups by their biological, physical and chemical characterisation. A higher-level grouping of the RIVPACS end groups into seven 'super groups', primarily for mapping and descriptive purposes, provides a broad classification of river typology (Table 5.4) (Davy-Bowker et al. 2008). Sites pertaining to the three super groups predominantly comprising lowland characteristics (E, F and G) were retained (end groups 31-43). The term 'intermediate' is used to qualitatively describe rivers in supergroups C and E, however this is not clearly defined in RIVPACS documentation. It is understood this phrase relates to the size of the river and intermediate rivers are those which are of 'medium size' (comparable to 'small' rivers described in supergroup D and 'large' rivers described in supergroup G) (John Murray-Bligh, Environment Agency Lead for Invertebrates on UKTAG Freshwater Task Team, *pers. comm*).

Table 5.4 - Super group classification of the 43 RIVPACS end groups (Davy-Bowker et al. 2008).

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#### Phase 3 – Habitat quality

Along with water quality, habitat quality is of equal importance in determining macroinvertebrate community structure. Anthropogenic physical changes to a river will inevitably affect the balance of erosion, transport and deposition of fine sediment. Sites with any capital works (*'the percentage of the length of the river* 

(*km*) with river channel works that have major and lasting impacts on the channel morphology e.g. bank reinforcement, re-sectioning, re-alignment and re-grading as well as embankments'; Lathouri and Klaar 2016, p. 6) or re-sectioning were removed from the sites list. This is based on previous work by Dunbar et al. (2010) that showed these variables as important drivers of habitat quality based on their interaction with flow.

## Phase 4 – Hydrology

There was no capacity to install hydrological monitoring equipment at each site. Thus, in order to gain matched hydrological data, each site needed to be associated with an operating flow gauging station. A list of gauging stations was obtained from the National River Flow Archive (NRA). ARCGIS software (ESRI 2011) was used to map the coordinates of flow gauging stations (obtained from the National River Flow Archive 2018) and the coordinates of the filtered biological monitoring sites. Using the buffer and crop tools in ArcGIS 2.7, all sites within 2km of a gauging station were retained and those that were not were removed from the list. These were then manually checked to ensure the gauging station and monitoring site were associated with the same water body.

#### Final site selection

The final number of sites left after the filtering process was 27 (see Appendix 3.2 for maps of each filtering process). The number of sites was reduced to 21 once accessibility was taken into consideration (i.e. public land or where landowner permission could be obtained) (Table 5.5). The final list of sites showed a multi-region distribution throughout lowland England, with a range of RIVPACS end groups represented (Figure 5.3a). In order to ensure that these sites covered a range of fine sediment conditions they were checked using the Agricultural Sediment Risk Ratings map (ASR) from Naura et al. (2016). Agriculture is the main source of fine sediment inputs to river systems, and the ASR combines sediment inputs from land-based models and predictions of fine sediment accumulation using RHS data. The ASR gives a risk category of 1-5 (very low to very high). The ASR scores were retrieved for each site which showed there was some variation in risk categories between sites (Figure 5.3b).

Field	End	Super				Mean	Mean	Sand, silt
site id	group	group	Easting	Northing	Town, County	(m)	(cm)	(%)
177	41	G	434,583	466,973	Harrogate, Yorkshire	4.8	86.7	87
1,313	32	E	413,981	438,225	Bradford, Yorkshire	30.7	29.44	28
7,694	35	E	378,630	167,000	Bathford, Somerset	6	34	10
8,614	39	F	382,550	114,664	Hammoon, Dorset	2.3	31.88	59
9,144	40	F	415,025	107,225	New Forest, Hampshire	3.5	14	7
10,533	35	E	320,680	125,010	Taunton, Somerset	7.3	22.67	38
35,479	39	F	416,366	222,549	Cotswolds, Gloucestershire	6.2	23.67	70
35,614	40	F	501,202	147,423	Guildford, Surrey	4.9	20	44
42,051	40	F	543,850	122,580	Wealden, East Sussex	2	28.33	34
42,744	35	E	513,411	133,381	Horsham, West Sussex	2.5	14.33	25
42,794	41	G	521,500	117,220	Horsham, West Sussex	3.6	31.78	57
43,795	40	F	551,460	136,870	Wealden, East Sussex	3.7	53.33	20
49,306	41	G	376,730	224,790	Newent, Gloucestershire	3.3	35	87
54,650	38	F	576,230	237,740	Braintree, Essex	1.7	12	55
65,511	32	E	361,688	469,995	Lancaster, Lancashire	13.7	20.33	0
67,895	38	F	350,137	417,399	Chorley, Lancashire	3	21.33	28
81,003	36	Е	603,200	235,500	Babergh, Suffolk	6	42.5	50
155,066	40	F	486,730	121,952	Chichester, West Sussex	4.7	33	70
161,030	37	F	388,405	285,925	Dudley, West Midlands	7	30	10
161,225	35	E	320,415	125,482	Taunton, Somerset	2.9	13.4	25
162,069	37	F	353,166	144,059	Mendip, Somerset	3.5	30	12

Table 5. 5 – List of sites sampled. Mean width, depth and percentage sand, silt and clay is derived from River Habitat Survey and is therefore a long-term average of the values for each site.



Figure 5.3 – Sites sampled colour coded by RIVPACS end group classification (a) and Agricultural Sediment Risk Rating ranging from 1 (green) low risk to 5 (red) high risk (b).

## 5.3.2 Field data collection

In order to take account of natural seasonal variation in life-history patterns of diverse macroinvertebrate species, standard national monitoring practice is to sample macroinvertebrate communities during spring and autumn and an average score is provided for environmental health assessments. The sampling was therefore carried out within the time frames set by the EA for seasonal sampling (spring March-May; autumn September-November). Sites were visited for autumn sampling between 11<sup>th</sup> September 2016 and 3<sup>rd</sup> October 2016 and spring sampling between 13<sup>th</sup> May 2017 and 30<sup>th</sup> May 2017. EA site location cards (providing maps, site pictures, access points and any relevant safety concerns) were obtained for all 21 sites visited (see Appendix 3.3 for an example). Where the exact monitoring site was either unclear or inaccessible, the nearest suitable reach was sampled. The sampling area was accessed from the downstream end where possible so as not to disturb the river bed. This enabled some standard environmental variables to be collected which could explain community response, e.g. shading, detritus etc. A 50 ml background

water sample was collected at each site in order to quantify the suspended sediment concentration (SSC mg I<sup>-1</sup>) at the time of sampling.

Two principle methods of measuring fine sediment were carried out at each site: the resuspension method and visual estimates. The resuspension method was carried out within the reach four times; twice at erosional areas and twice at depositional areas (Figure 5.4). The method outlined in Duerdoth et al. (2015) was followed: an open-ended hollow cylinder of 0.56 m diameter was pushed into the gravel bed to achieve an adequate seal from the surrounding flow. Once a seal was achieved, three depths at random locations within the cylinder were taken using a metre rule and the average depth of water recorded. The water within the cylinder was then vigorously agitated for 60 seconds without touching the river bed in order to bring loose overlying sediment into suspension and the overlaying water was sampled. An electric drill with plaster mixing attachment (Figure 5.5a) was used for this in order to standardise the mixing. Immediately following the 60 second agitation, a water sample was taken by pushing an inverted 50 ml measuring cylinder into the middle of the water column within the cylinder and turned upright so it filled as it was drawn to the surface in order to collect a well-mixed sample (Figure 5.5b). The cylinder was emptied into a clean sample bottle and rinsed with clear river water into the sample bottle to ensure no residue was left in the cylinder. The process was then repeated with 30 seconds of subsurface agitation using a metal auger to raise subsurface fine sediment into suspension (Figure 5.5c), then 30 seconds of overlying water agitation using the electric drill with mixing attachment. The subsurface agitation aims to disturb the top 100 mm of the gravel bed. A further water sample was then taken to characterise the total fine sediment (from the subsurface agitation which ultimately includes both surface and subsurface fine sediment). The calculations to convert the mass of fine sediment extracted from the sample volume within the cylinder to g m<sup>-2</sup> are summarised in Equations 5.1 - 5.3. All water samples (including the 50 ml background SSC sample) were kept in a cool box with ice during field work and then transferred to a fridge in the laboratory on return.


Figure 5.4 – An example sediment sampling layout per reach. The resuspension samples are labelled ascending upstream demonstrating the direction of sampling.

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Figure 5.5 – Use of the cordless electric drill with plaster mixing attachment (a), collecting water samples after agitation from the bottom of the cylinder (b) and agitating the subsurface using a metal auger (c).

(Dry weight of filter (g) + filtrate(g)) - Dry weight of filter paper (g) (Eq 5.1)= Total mass in sampled volume (g)

 $Total mass in sampled volume (g) \times \frac{Total volume in cylinder (ml)}{Filtered volume (ml)}$ (Eq 5.2) = Total mass in stilling well (g)

$$\frac{1 (m^2)}{\text{Area of stilling well } (m^2)} \times \text{Total mass in stilling well } (g)$$

$$= \text{Total mass per } g m^{-2}$$

Ideally, there would be clear erosional (e.g. riffle) and depositional (e.g. pool) areas at each site. However, not all sites have a clear distinction and some lacked a clearly defined pool-riffle structure. The areas were chosen to be representative of the site. Duerdoth et al. (2015) suggests that as fine sediment appears to have a highly skewed distribution in rivers (most of the fine sediment is confined to patches and the majority of the riverbed contains little sediment), it is better to sample the extremes of the distribution and calculate the geometric mean as an estimate of central tendency, rather than an arithmetic average of random samples (as done by Clapcott et al. 2011). It was noted that the method did not work particularly well in coarse substrates or sites with a predominantly bedrock substrate because it was difficult to obtain a seal with the bed which resulted in water movement through the cylinder and winnowing of fine sediments out of the cylinder. This reduces the concentration of fine sediment within the cylinder and would result in underestimating the mass of fine sediment stored in each reach. It was also noted that when attempting to achieve a seal with the substrate the operator can introduce bias by selecting areas of the bed which are less coarse and a seal is more readily achievable. This could result in overestimating the concentrations of fine sediment within a reach.

Visual estimates of fine sediment were taken at the reach scale (Figure 5.4). As described in the River Habitat Survey Field Survey Guidance Manual (Environment Agency 2003), visual estimates involve the operator estimating the percentage substratum composition over a given reach. Substrate categories comprised; bedrock, boulders (>256 mm), cobbles (64 - 256 mm), pebbles (4 - 64 mm), gravel (2 - 4 mm), sand (0.0625 - 2 mm), silt (<0.0625 mm) and clay (cohesive). The reach scale visual estimates were made by walking up the length of the reach on the river bank, and also by entering the reach to confirm substrate type, and recorded (Appendix 3.4). Visual estimates were also taken at the patch scale within the resuspension cylinder before any agitation had occurred to allow comparisons between the quantitative and semi-quantitative methods.

The biological sampling method used to collect macroinvertebrates was the semi-quantitative multi-habitat three-minute survey using a standard kick net protocol (Friberg et al. 2006, Environment Agency 2014a) (Figure 5.6). A single three-minute survey was carried out at each site (for each season). Sampling started at the most downstream area with the operator kicking to dislodge the substratum up to a depth of 100 mm whilst holding a long-handled pond net at a right angle to the current to collect any invertebrates that have been disturbed. The operator moved upstream in a diagonal motion across the channel, sampling continuously (i.e. not in patches or bursts). The total sampling time was distributed proportionally between the habitats present in the sampling area. For example, if 50% of the sampling reach was a riffle habitat then 50% of the time was spent sampling across riffle areas. This method is ineffective in sites where the stream bed is completely covered in silt as the net clogged guickly. Instead, in these areas the net was gently skimmed through the top layers of sediment. The kicking procedure was followed by a one-minute manual search of the sampling area to determine surface dwelling organisms (e.g. pond skaters) or attached animals (e.g. caddis pupae or leeches) to logs, stones, overhanging vegetation or other solid objects. This aims to cover what is missed using the kick-net sampling method. Following collection, the contents of the pond net were transferred to a plastic container and filled with 70%

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Industrial Methylated Spirits (IMS) to preserve the sample (Environment Agency 2015).





At each site, three measurements of the wetted channel width, using a field tape measure, were made along the reach. An average of the depths taken for the resuspension cylinders was taken for the reach-scale water depth. To minimise sampling error, the same operator was used for all sample collection, i.e. surface and subsurface agitation, resuspension sample collection, background sample collection, visual estimates of fine sediment and the kick net sampling. A second operator was always present during field work for equipment assistance and safety.

# 5.3.3 Laboratory methods

The refrigerated water samples collected from the resuspension method were processed within four days of collection. The processing method used followed that of Duerdoth et al. (2015). The sample bottles used for the 50 ml volume collection were removed from the fridge and poured through a 2 mm sieve onto a 90 mm GF/C Whatman glass microfibre filter paper. Filter papers were pre-

ashed (at 500 °C for 2 hours) and washed in deionised water prior to use in order to remove any contaminants left on the filter papers during the manufacturing process. The filter papers were weighed on a micro-balance to 0.00001 g. During a pilot study, it was found that a significant amount of sediment was left around the filter holder after filtering despite rinsing with reverse osmosis deionised (RO DI) water. Therefore, all filtration was carried out using gravity filtration only; a folded filter paper was placed inside a plastic funnel balanced over a conical flask to collect the filtrate. A wash bottle filled with deionised water was used to rinse the collection bottle into the filter paper to collect any residue. The filter papers were dried overnight in an oven at 105 °C and cooled in a desiccator for 30 minutes before weighing to determine total mass of sediment retained. The filter papers were ignited in a furnace at 500 °C for 30 minutes and again cooled in a desiccator before weighing to determine the mass of organic matter lost through ignition (LOI).

The factors affecting LOI, as identified in the well-cited paper on LOI for estimating organic carbon content in sediments by Heiri, Lotter, and Lemcke (2001), were acknowledged. Three recommendations for consistency in LOI analysis were considered: ignition temperatures, exposure times and sample size. The temperature, duration of ignition and sample size used during this study was selected as recommended by Duerdoth et al. (2015) and has since been used by several other studies (Naden et al. 2016, Conroy et al. 2016b) and therefore a method standard has already been established. By using the same parameters, the results of the present study can be comparable to these earlier studies. In order to minimise error, the same balance was used for all laboratory processes. Due to the very small mass of fine sediment collected from a 50 ml sample, it was sometimes found that the mass of the filter paper after ignition was less than the mass at the beginning. Therefore, filter paper 'blanks' were also analysed by following the same process, filtering only deionised water and calculating the mass loss of filter paper through the process which was then used for correcting the mass of sediment. It is recommended that a larger volume of water is taken for future analytical methods to help overcome this variation.

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Macroinvertebrate samples were processed following EA protocols (Environment Agency 2014b). The samples were emptied into a 500 µm sieve and rinsed under a fume hood to remove the IMS. Once the IMS had been removed, the sample was thoroughly rinsed to remove excess silt. After the silt had been removed, about one tablespoon of the sample was removed at a time, placed in a white sorting tray, submerged in water and observed under a magnifying lamp. The sorting tray was divided into grid sections using a permanent marker to allow sorting in a systematic way, working through the tray contents section by section rather than at random. Taxon identification was conducted under a high-powered light microscope. Macroinvertebrate samples were identified to mixed taxon level following the standard operating procedure of the (Environment Agency 2014b). At mixed taxon level, most insect taxa are identified to species level with the exception of some Diptera families (see Appendix 3.5).

## 5.3.4 Data analysis

#### 5.3.4.1 Calculating sediment metrics

The SSC for each site was calculated from the background sediment samples (mg l<sup>-1</sup>). Processing the surface agitation resuspension samples yielded the following metrics: total surface sediment (g m<sup>-2</sup>), total organic surface sediment (g m<sup>-2</sup>), total inorganic surface sediment (g m<sup>-2</sup>). Processing the subsurface agitation samples yielded the following metrics total sediment (g m<sup>-2</sup>), total organic sediment (g m<sup>-2</sup>), and total inorganic sediment (g m<sup>-2</sup>). As the subsurface agitation incorporates both the surface sediment and the sediment from the top 100 mm of gravel, these metrics are described as the 'total' sediment. Following the methods as set out in Duerdoth et al. (2015), the geometric mean of the data for each of the four samples at each site (two erosional and two depositional) was calculated providing a single figure for each of the measures for each site. Resuspension samples were corrected for background SSC. For both the SSC and the metrics derived from the resuspension sampling regime, a further correction was applied based on the average mass loss of filter paper fibres during the filtering, drying and firing

processes (see description of 'blanks' in Section 5.3.3). Following this correction, any measures which still remained negative were corrected to zero.

To calculate the percentage of reach scale visual fines for each site, the sum of the estimated clay, silt and sand fraction were combined. Patch scale estimates were calculated using the same aggregation of substrates using the visual estimates from within the resuspension cylinder before agitation. Patch scale estimates are specified where included in the data analysis. However, as the most relevant method to the optimisation of EPSI and EPSImixed, the reach estimates of visual fines are predominantly used throughout.

### 5.3.4.2 Hydrological metrics

Mean daily flow (discharge m<sup>3</sup> s<sup>-1</sup>) was obtained for each site for the period 01/01/2000 – 31/05/2017. Missing data were imputed using the *missForest* package (Stekhoven and Buhlmann 2012). The *missForest* function uses random forests trained on the observed values to predict the missing values. The 'out of bag' errors (a measure of cross-validation), presented as the normalized root mean square error (NRMSE) for continuous variables, compares the observed data with the imputed (full) data matrix. The NRMSE for the whole imputation was 0.06 (i.e. the variables are imputed with 6% error). There is no pre-determined acceptable value for NMRSE, however lower values (closer to zero) represent more robust imputations. The NRMSE for this imputation is determined acceptable.

Two sets of hydrological metrics were calculated from the data to describe (a) the flow regime and (b) the antecedent flow. Flow data were standardized prior to analysis (using the *scale* function in R). Following standard practice (e.g. Mathers 2017), standardization was carried out by first centering by the mean and then dividing by the standard deviation to convert the data to Z-scores. This enables comparison between sites as flow will inherently vary as a function of site. The flow regime metrics were based around the five critical components of the natural flow regime as outlined by Poff et al. (1997): magnitude, frequency, duration, timing and rate of change. In total, 22 flow regime metrics (Table 5.6) were calculated based around these five facets and identified from previous

studies reporting that these metrics are closely related to ecological structure and function (Olden and Poff 2003, Monk et al. 2007). Ninety-six metrics were adopted to describe the antecedent flow conditions (Table 5.7). Lastly, stream power was calculated using the formula  $\Omega = \rho g Q S$ . where  $\rho$  is the density of water (1000 kg m<sup>3</sup>), *g* is acceleration due to gravity (9.8 m s<sup>2</sup>), *Q* is the mean daily discharge calculated from the average mean daily discharge for the entire data period for each site (m<sup>3</sup> s<sup>-1</sup>), and *S* is the channel slope at each site was calculated.

Flow regime metrics	Description
TOTALVOL	Total discharge for year to date
MDF	Mean daily discharge (for entire time series)
MADQ	Mean annual discharge
DAY90MAX	Average annual maximum 90-day discharge
DAY30MAX	Average annual maximum 30-day discharge
DAY7MAX	Average annual maximum 7-day discharge
MMAD	Maximum annual monthly discharge
DFMEDMAX	Median of the maximum annual monthly discharge/median annual daily discharge
STDEVDF	Standard deviation of the daily discharge
DFQ95MEAN	Q95/MDF
BASEFLOW	7-day annual minimum discharge/MADQ
DFBFI	Mean of lowest annual daily Q/mean of lowest annual daily Q
Q1090DF	Q10/Q90
CVANNQ	Covariance of MADQ
FRE1YR	Mean number of events per year over Q50
SK2	(MADQ – median annual Q)/median annual Q
Q550DF	Q5/Q50
Q10DF, Q25DF, Q20DF, Q5DF, Q1DF	The flow that is exceeded for a given percentile of time
StreamPower	Calculated as $\Omega = \rho g Q S$ for the entire data period for each site

Table 5.6 – Hydrological regime metrics calculated from daily discharge data for all sites.

Antocodont				Description	
flow metrics	Description		Time frames	(all relative to sampling date)	
MDF	Mean daily discharge		Pre7d	Previous 7 days	
MAX	Maxima		Pre30d	Previous 30 days	
MIN	Minima		Pre6m	Previous 6 months	
SD	Standard deviation		Pre12m	Previous 12 months	
		+	PreSum	Previous summer (June, July & August)	
Q1 Q5 Q10	The flow that is		PreSpr	Previous spring (March, April & May)	
Q20 Q25 Q50 Q90	Q10The flow that is exceeded for a given percentile of timeQ25given percentile of timeQ50Q90Q95Q95		PreAut	Previous autumn (September, October & November)	
Q95			PreWin	Previous winter (December, January & February)	

Table 5.7 – Antecedent flow metrics. Each metric (left) was calculated for each of the time frames (right) prior to each sampling date e.g. MDFPre7d.

When calculating a large number of hydrological metrics for both flow regime and antecedent flow, there is a high degree of redundancy. In order to reduce redundancy, existing methods developed in ecohydrology were applied (e.g. Olden and Poff 2003; Monk et al. 2007; White et al. 2017). Principal Component Analysis (PCA) (using the function *prcomp* in R) was calculated on each of the sets of indices individually. All statistical analysis was carried out using R version 3.5.3 (R Development Core Team 2019). The purpose of PCA is to reduce dimensionality whilst still preserving variance (Jollife and Cadima 2016) and is therefore a common method in dimensionality reduction. Unlike linear regression, PCA models are not destabilised by collinearity between variables. However, similar to linear models, PCA relies assumes a normal distribution of the data. The first two principal components (PC) contributed 92.08 % to the total variance for the flow regime indices and 82.47 % for the antecedent flow indices. Since there was a high amount of collinearity for both sets (Appendix 3.6) the 'broken stick' method was used to select non-collinear variables (Olden and Poff 2003) which is described as follows. The contribution of each of the variables to dimensions 1 and 2 (in descending order) were calculated (Appendix 3.6). The correlation coefficients of the indices were calculated using Pearson's product moment correlation (*cor* function in R). Forward selection was carried out so that the metric contributing most to the first two PCs was retained if the Pearson's correlation coefficient (*r*) between any pair of variables was higher than 0.95 (the number at which the relationship is deemed to be perfectly collinear; White et al. 2017). PCA plots for the retained set of metrics are shown in Figure 5.7 and 5.8.



Figure 5.7 – Principal Component Analysis for flow regime metrics retained after removing redundant metrics.



Figure 5.8 - Principal Component Analysis plots for antecedent flow metrics retained after removing redundant metrics.

## 5.3.4.3 Methods of measuring fine sediment

Early data visualisation of the variation in environmental variables between sites was carried out using PCA (using the *prcomp* function in R). Spearman's rank correlation was used to compare the different metrics of fine sediment (using *cor* function) as the data were not-normally distributed (confirmed by *shapiro.test* function with p values <0.05). A model selection process using both linear modelling (*Im* in R) and mixed effects modelling (*Imer* in R; fitted using maximum likelihood estimation) was used to determine whether season had a significant effect on the relationship between the semi-quantitative estimates of fine sediment (derived from visual estimates) and the fully-quantitative total surface sediment and total subsurface sediment (derived from the resuspension

sampling). The response variables were log(x+1) transformed to reduce skewness (observed from histograms). The optimal models were determined as the most parsimonious model with the lowest Akaike's Information Criterion (AIC) value, or the next lowest if the difference was <2 AIC points (Burnham and Anderson 2004). In the case of both total surface and total sediment, the optimal model did not include season as either a fixed or random effect (Table 5.8).

Table 5.8 - Model selection method process predicting total surface and total sediment from visual fines based on season. The optimal models are marked with an asterisk.

Model	AIC				
MOUEI	Total surface	Total sediment			
Total surface ~ visual fines	136.967*	144.869*			
Total surface ~ visual fines + season	138.848	145.888			
Total surface ~ visual fines + (1 season)	138.967	146.869			
Total surface ~ visual fines + season + (1 season)	140.848	147.888			

Linear modelling was also used to determine which environmental variables affect each metric of fine sediment. The retained hydrological metrics after the variable reduction procedure were combined with environmental data from the field sheet (Appendix 3.4) to derive a full list of predictors. Categorical variables from the field sheet were converted to numerical values for analysis. In order to incorporate variations between the catchment land use at each site, the Agricultural Sediment Loading (ASL) index was included as a predictor (Naura et al. 2016). The ASL provides an estimate of the quantity of fine sediment from agricultural origin delivered to reaches through run off. The index is derived from GIS mapping processes through the Phosphorous and Sediment Yield CHaracterisation In Catchments model (PSYCHIC). The ASL provides one half of the contribution towards the Agricultural Sediment Risk (ASR) index which was used in the earlier site filtering process (Section 5.3.4.1).

Because of the high number of predictors, and the risk of overfitting in the modelling process, the variance inflation factor (VIF; using *corvif* function in R) was used to reduce the number of predictors based on their collinearity. Forward stepwise selection was carried out, the predictor with the highest VIF removed and the function run again. The recommendation given by Zuur et al. (2009) is to remove variables until all VIF values are below 3 or 5. The higher value of 5 was chosen here due to the risk of excluding ecologically relevant variables with the more stringent threshold. A full list of the original predictors and the refined list after the VIF analysis was carried out can be found in Appendix 3.7.

The fine sediment metrics were log or log(x+1) transformed prior to modelling to reduce skewness (observed from histograms). Model selection was carried out to determine whether season should be included as a fixed effect, random effect or both (Table 5.9). As before, the optimal models were determined as the most parsimonious model with the lowest Akaike's Information Criterion (AIC) value, or the next lowest if the difference was <2 AIC points (Burnham and Anderson 2004). Stepwise selection was used to reduce the optimal models for each metric (using the *StepAIC* function in R, direction = 'both'). Earlier analyses showed a relatively strong fit among the deposited metrics of fine sediment. As the aim of this specific analysis was to determine which environmental variables affect each metric of fine sediment, the deposited metrics were not included as predictors for these sets of models. Suspended sediment appears independent of deposited sediment and therefore background SSC was offered as a predictor for each deposited sediment model.

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Table 5.9 – Model selection process for each metric of fine sediment. Optimal models are marked with an asterisk.

Model	environmental variables as fixed predictors	environmental variables as fixed predictors + season	environmental variables as fixed predictors + season + (1 season)	environmental variables as fixed predictors + (1 season)
Visual fines	64.352*	66.289	68.352	66.352
Background SSC	139.086	136.544*	138.544	140.831
Total surface	145.486*	144.557	146.557	147.486
Organic surface	119.052	110.554*	112.554	117.122
Inorganic surface	140.527*	141.596	143.596	142.527
Total sediment	126.832	123.969*	125.969	128.551
Total organic	138.878	134.575*	136.575	139.857
Total inorganic	137.824*	136.699	138.699	139.824

5.3.4.4 Assessing the performance of sediment-specific biomonitoring indices

A complete list of the observed taxa was compiled for each site. As stated in Section 5.3.3, taxa were identified to the lowest taxonomic resolution possible. However, for particularly small or damaged individuals the taxa were taken to a coarser level (e.g. genus or family level). A set of rules (below) was developed in order to assign a biotic index score to taxa identified at a coarser taxonomic level (e.g. genus and family) than scored under the index (e.g. species level). The set of rules were developed with advice from Richard Chadd (Taxonomic Lead for the Environment Agency). It is recognised that some families/genera will contain species scores across a wide range of sensitivities, reflecting response diversity within the taxonomic rank (e.g. Limnephilidae). However, in these instances, the resulting score derived through the averaging process often approached a median value (of the index range) and was therefore noninfluential on the calculation of the overall score for each site. The set of rules were as follows:

- Where a genus level does not score, the average from the species scoring within that genus is used (for PSI, which has a categorical scoring system, the mode was used in the first instance and then median for PSI if there was no mode)
- Where a family level does not score, the average from all species scoring within that family is used (as above for PSI)
- For a species that does not score but there is a genus level score, this score is given to the species
- Additionally, all species/genera are included for the purposes of averaging regardless of ubiquity and prior knowledge (e.g. rare species are included in the averaging process)

The fuzzy coded Tachet et al. (2010) trait database was used to assign trait scores to each taxon. This particular trait database was chosen as it is one of the most comprehensive databases and has been widely used in ecological studies. Only true traits, and not preferences, were assigned to each taxon (Violle et al. 2007). As with other biotic indices, in instances where index scores were available at a coarser taxonomic level than traits, fuzzy values were averaged across genera or species. The complete trait dataset comprised 63 trait modalities in 11 trait categories (Table 5.10). Functional trait diversity was calculated using the dbFD function in the FD package in R (Laliberté, Legendre, and Shipley 2014). Prior to calculating functional diversity (FD), the traits were converted to proportions (i.e. removing fuzzy coding) and centred but not standardized (Chevene, Doledec, and Chessel 1994). The dbFD function implements a distance-based framework to compute multidimensional functional indices (Laliberté, Legendre, and Shipley 2014). The FD indices calculated were functional richness (FRic; Villéger, Mason and Mouillot 2008), functional dispersion (FDis; Laliberté and Legendre 2010) and the communitylevel weighted means of trait values for shredders (CWM; Lavorel et al. 2007).

The CWMshredders was chosen as existing literature has pointed to shredders showing a particular sensitivity to fine sediment (see Chapter 3). Burrowers were also been identified as consistently tolerant to fine sediment. However, the relationship is potentially more complex as burrowers can also be sensitive to fine sediment depending on the substrate they burrow into (Wilkes et al. 2017). Therefore, only CWMshredders was calculated. As well as the sediment-specific biomonitoring indices and FD indices, indices used in national biomonitoring practices were calculated (Table 5.11). For indices scored at family level, the biotic package in R was used (Briers 2016).

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Table 5.10 – Trait categories, modalities and short names used for analysis (from Tachet et al. 2010).

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Index	Short name
Proportion of Sediment-sensitive Invertebrates	PSI
Empirical Proportion of Sediment-sensitive Invertebrates	EPSI
Empirical Proportion of Sediment-sensitive Invertebrates family level	EPSImixed
Combined Fine Sediment Index	CoFSI
Organic Fine Sediment Index (constituent of CoFSI)	oFSI
Total Fine Sediment Index (constituent of CoFSI)	ToFSI
Wally Hawks Paisley Trigg Index – average score per taxon	WHPT_ASPT
Wally Hawks Paisley Trigg Index – number of scoring taxa	WHPT_NTAXA
British biomonitoring Working Party score	BMWP
British biomonitoring working party - average score per taxon	BMWP_ASPT
British biomonitoring working party - number of scoring taxa	BMWP_NTAXA
Lotic Index Flow Evaluation	LIFE
Ephemeroptera, Plecoptera, Trichoptera (richness)	EPT
Abundance	Abundance
Shannon's Diversity	Shannons
Functional richness	FRic
Functional dispersion	FDis
Percentage of shredder taxa	Shredderpercentage
Community weighted means (extracted from dbfd) of shredders	CWMshredder

Table 5.11 – A full list of macroinvertebrate indices calculated for analysis.

Spearman's rank correlation was used to assess the performance of the indices against fine sediment metrics. The data were tested for normality using the Shapiro-Wilk test which confirmed that the distributions of the index scores were significantly different from the normal distribution, justifying the use of the nonparametric test. As the pairwise correlations were calculated at one time, the Holm-Bonferronni correction (Holm 1979) was applied to reduce the chance of type 1 errors (false rejection of the null hypothesis i.e. a false positive). Linear modelling was used to determine the variables affecting index performance. The same process outlined in Section 5.3.4.3 was used to reduce the VIF of the variables offered to the model (a full list of the variables offered to the model can be found in Appendix 3.8). Model selection was carried out to determine whether season should be included as a fixed effect, random effect or both. Data (predictor variables) were scaled prior to the model selection process (using the scale function in R). Results of the model selection process can be found in Appendix 3.9. For PSI, oFSI, ToFSI, WHPT\_ASPT, LIFE, EPT and FRic, the optimal model included season as a fixed effect. For EPSI, EPSImixed, CoFSI, WHPT\_NTAXA and FDis, season was not included in the model. Stepwise selection was used to refine the optimal models for each metric (using the StepAIC function in R, direction = 'both').

5.3.4.5 Assessing indicator taxa, traits and trait-environment relationships

One of the objectives of this chapter is to use a range of robust statistical methods in order to improve our understanding of the response of macroinvertebrates to fine sediment pollution. Firstly, Threshold Indicator TAxa ANalysis (TITAN) (Baker and King 2013) was applied to visual fines, total surface, organic surface, and total sediment (*titan* function in the *TITAN2* package in R; Baker, King, and Kahle 2015). TITAN uses a resampling technique to detect taxon-specific changepoints of abundance and occurrence across an environmental gradient. In order to calculate TITAN, taxa occurring at less than three sites must be removed (full list of the 106 taxa included in the TITAN analysis can be found in Appendix 3.10). TITAN analysis was carried out using both taxonomic (absolute abundance) and trait (CWMs) data for the

different metrics of fine sediment. The parameters in the TITAN function were set as 1000 random permutations (*numPerm*) and 1000 bootstrap replicates (*nBoot*). These parameters were selected based on recommendations by Baker, King, and Kahle (2015) for formal analysis.

Fourth corner and RLQ analysis (using *fourthcorner, fourthcorner*2 and *rlq* functions in the *ade4* package in R; Dray et al. 2018) was carried out to test for links between traits, taxa and environmental variables across all sites (Verberk, van Noordwijk, and Hildrew 2013). Gradient forest (GF) (using the *gradientForest* package in R; Ellis, Smith, and Pitcher 2012) was used to compare the turnover of taxonomic and trait structure across visual fines and total surface sediment to identify the taxa and traits most susceptible to changes in abundance across the gradients. The importance of each environmental variable in determining the taxa and trait turnover was extracted from the GF models. The reduced set of variables used in biomonitoring index modelling (listed in Appendix 3.8), with all sediment variables included, was used for both the RLQ and the GF.

# 5.4 Results

# 5.4.1 Data summary

Full results for visual assessments of fine sediment and metrics derived from the resuspension method for each site can be found in Appendix 3.12. The first two PCs contributed 49.2% of the total explained variance (see Appendix 3.11 for scree plot). Spring and autumn site data were well integrated and did not form distinct groups in the ordination plot (Figure 5.9). The top variables contributing most to the primary PC were mostly sediment metrics whereas other physical habitat parameters contributed most to PC2 (Figure 5.10). This confirms that the sampling regime captured a habitat gradient dominated by fine sediment conditions.



Figure 5.9 – Principal Component Analysis of the environmental data, plots showing as a variable plot (a), individual sites labelled by seasons (b).



Figure 5.10 - Top 10 variables contributing to PC1 (a) and PC2 (b).

During the site filtering process, the ASR was used to determine whether there was a gradient of fine sediment across each site. The ASR category for each site was compared to the quantitative physical measurements of fine sediment (Figure 5.11). It is difficult to assess whether there is broad agreement between quantitative measures of fine sediment with the ASR ratings as there were not enough sites at the higher risk categories.



Figure 5.11 – Scatterplot of sites by agricultural sediment risk group (ASR) and total surface sediment (g  $m^{-2}$ ).

### 5.4.2 Comparing methods of measuring fine sediment

There was a strong correlation between reach scale visual estimates of fine sediment and total surface sediment ( $\rho = 0.82$ , p <0.001). The relationship was stronger at the patch scale ( $\rho = 0.90$ , p <0.001) (Figure 5.12). Visual fines, at both the reach and patch scales, correlated less well with organic metrics (organic surface  $\rho = 0.53$ , p = 0.029, total organic  $\rho = 0.62$ , p <0.001) than inorganic metrics (inorganic surface  $\rho = 0.82$ , p <0.001, total inorganics  $\rho = 0.73$ , p <0.001). There were strong and significant correlations between most of the metrics derived from the resuspension method with the exception of organic surface sediment which was weaker, albeit still significant. Notably, the correlation between organic surface sediment and total surface sediment was weaker ( $\rho = 0.65$ , p <0.001) compared to the almost perfect correlation of total surface sediment with inorganic surface sediment ( $\rho = 0.99$ , p <0.001). SSC levels were not significantly correlated with any deposited metrics.



Figure 5.12 – Spearman's rank correlation matrix of metrics of fine sediment. Font size of the correlation coefficient is scaled to coefficient value. Significant correlations are marked with an asterisk.

The correlation between visual estimates and total surface sediment was stronger for spring ( $\rho = 0.879$ , p <0.001) than autumn ( $\rho = 0.762$ , p <0.001). However, model selection determined that the linear model without season as either a fixed or random effect was optimal for both total surface and total sediment (Table 5.8). Both models were significant with the model fit (R<sup>2</sup>) of total surface higher than total sediment (Table 5.12).

Model	Coefficient	Estimate	Std.	t	р
			Error	value	
Total surface ~ visual	Intercept	2.141	0.297	7.199	<0.001*
fines					
<b>df</b> 40					
<b>Adj R</b> <sup>2</sup> 0.556	Visual fines	0.048	0.007	7.23	<0.001*
<b>F</b> 52.32					
<b>p</b> <0.001*					
Total sediment ~	Intercept	3.560	0.327	10.894	<0.001*
visual fines					
<b>df</b> 40					
<b>Adj R<sup>2</sup></b> 0.420	Visual fines	0.040	0.007	5.537	<0.001*
<b>F</b> 30.66					
<b>p</b> <0.001*					

Table 5.12 – Linear mixed effect model results predicting total surface sediment from visual fines. Significant coefficients are marked with an asterisk.

When determining the significant environmental predictors of each fine sediment metric, model selection determined that the linear model with season included as a fixed effect was optimal for organic surface, total sediment, total organic and background SSC (Table 5.9). This is intuitive, at least for the organic metrics, due to seasonal changes in organic inputs. Season was not included as a fixed effect for the remaining sediment metrics. Diagnostic plots for all models can be found in Appendix 3.13. All models were significant (Table

5.13 - 5.16), and the adjusted  $R^2$  was particularly high for all deposited metrics of fine sediment, with the exception of total surface sediment for which it was more moderate ( $R^2 = 0.66$ , p < 0.001). The adjusted  $R^2$  was relatively low for background SSC ( $R^2 = 0.3$ , p = 0.011). Model results for organic surface, inorganic surface, total organic and total inorganic can be found in Appendix 3.14. Width was a significant predictor, with a negative coefficient estimate (i.e. as width increases, the estimates of fine sediment decrease), for all metrics except organic surface and total organic. The coarse bed matrix (combined percentage of boulders, cobbles and pebbles) was significant for the metrics assessing deposited sediment, but not for background SSC. Season was significant for the metrics where it was included as a fixed effect (including the organic metrics). The high regime flow metric, Q1, and the relatively high antecedent metric, Q20pre7d, were either not retained or not significant for all sediment metrics except organic surface and total organics (where Q1 was significant) (see Table 5.6 – 5.7 for acronyms). The hydrological metric Q1090DF was significant for all metrics except total sediment. Notably, the coefficient was negative for background SSC but positive for all other deposited metrics. The antecedent flow metrics Q50preSum was significant for visual fines and total sediment, and Q50preWin was significant for visual fines only. The relatively high antecedent flow, Q20pre6m, was only significant for background SSC.

### 5.4.3 Testing the performance of sediment-specific biomonitoring indices

The observed PSI and EPSI index scores covered almost the full range of the index (PSI 7.14% - 86.67% and EPSI 16.33 – 95.21%). However, most sites score towards the upper end of the index, indicating that they were not particularly impacted by fine sediment (Figures 5.13 - 5.15). The CoFSI index score usually ranges from 3.0 - 6.5. The scores for the sites fell within this range (3.89 - 5.01). There appeared to be a negative relationship between the fine sediment metrics with both PSI, EPSI and CoFSI. WHPT\_ASPT appeared not to show any clear relationship with the fine sediment metrics.

Response	Coefficient	Estimate	Std. Error	t value	р
	(Intercept)	7.586	0.926	8.193	<0.001*
	Width	-0.075	0.021	-3.618	0.001*
	Bedrock	-0.009	0.007	-1.352	0.187
Visual fines	Macrophyte	0.363	0.121	3.008	0.005*
46.00	Altitude	-0.011	0.003	-4.136	<0.001*
	Slope	0.067	0.054	1.229	0.229
<b>AUJ K</b> $-0.862$	Background SSC	0.009	0.004	2.596	0.014*
$\Gamma 24.22$	Coarse bed matrix	-0.022	0.003	-6.656	<0.001*
<b>p</b> < 0.001	Q1	-0.246	0.122	-2.015	0.053
	Q1090DF	1.146	0.336	3.405	0.002*
	Q50preWin	0.669	0.229	2.924	0.007*
	Q50preSum	1.338	0.613	2.183	0.037*

Table 5.13 – Refined linear model results for visual fines. Significant effects are marked with an asterisk.

Table 5.14 – Refined linear model results for total surface sediment. Significant effects are marked with an asterisk.

Response	Coefficient	Estimate	Std. Error	t value	р
	(Intercept)	11.437	2.668	4.286	<0.001*
Teteleumfees	Width	-0.122	0.051	-2.379	0.023*
l otal surface	Depth	0.029	0.018	1.657	0.107
df oo	Bedrock	-0.033	0.018	-1.773	0.086
	Coarse bed matrix	-0.018	0.008	-2.069	0.047*
<b>AUJ K</b> $^{-}$ 0.002	Erosional flow	-0.008	0.006	-1.534	0.135
<b>r</b> 9.900	Q1	-0.463	0.308	-1.501	0.143
<b>p</b> <0.001	Q1090DF	1.806	0.811	2.228	0.033*
	Q50preSum	3.456	1.430	2.416	0.022*
	Stream power	0.346	0.200	1.731	0.093

Response	Coefficient	Estimate	Std. Error	t value	Р
	(Intercept)	8.606	0.897	9.590	<0.001*
	Width	-0.092	0.038	-2.418	0.022*
Total sediment	Bedrock	-0.070	0.013	-5.355	<0.001*
(subsurface)	Filamentous algae	0.292	0.168	1.741	0.092
	Macrophyte	0.379	0.216	1.752	0.090
<b>df</b> 31	Background SSC	0.015	0.007	2.081	0.046*
<b>Adj R<sup>2</sup> 0.779</b>	Coarse bed matrix	-0.030	0.007	-4.559	<0.001*
<b>F</b> 15.41	Erosional flow	-0.009	0.004	-2.338	0.026*
<b>P</b> <0.001*	Q1090DF	1.027	0.530	1.938	0.062
	Q20pre7d	1.397	0.725	1.928	0.063
	Season (Spring)	0.577	0.265	2.173	0.038*

Table 5.15 – Refined linear model results for total sediment. Significant effects are marked with an asterisk.

Table 5.16 – Refined linear model results for background SSC. Significant effects are marked with an asterisk.

Response	Coefficient	Estimate	Std. Error	t value	Р
	(Intercept)	-2.122	1.243	-1.707	0.097
Destaura	Width	-0.099	0.035	-2.825	0.008*
SSC SSC	Depth	0.050	0.014	3.460	0.002*
	Filamentous algae	0.550	0.181	3.036	0.005*
<b>df</b> 32	Macrophyte	-0.350	0.249	-1.405	0.170
<b>Adj R²</b> 0.302	Altitude	0.009	0.006	1.432	0.162
<b>F</b> 2.971	Erosional flow	0.009	0.005	1.865	0.071
<b>p</b> 0.011*	Q1090DF	-1.588	0.624	-2.543	0.016*
	Q20pre6m	-1.830	0.719	-2.546	0.016*
	Season (spring)	0.762	0.322	2.369	0.024*



Figure 5.13 – Plots of visual fines (%) with PSI (a), EPSI (b), CoFSI (c) and WHPT\_ASPT (d).



Figure 5.14 – Plots of total surface sediment (g  $m^{-2}$ ) with PSI (a), EPSI (b), CoFSI (c) and WHPT\_ASPT (d).



Figure 5.15 - Plots of total sediment (g m<sup>-2</sup>) with PSI (a), EPSI (b), CoFSI (c) and WHPT\_ASPT (d).

The strongest correlation was between visual fines and EPSImixed ( $\rho = -0.65$ , p < 0.001) followed by visual fines and EPSI ( $\rho = -0.60$ , p = 0.003) (Figure 5.16). Total surface sediment correlates most strongly with EPSImixed ( $\rho = -0.59$ , p = 0.006). CoFSI correlated significantly with both visual fines ( $\rho = -0.54$ , p = 0.022) and total surface sediment ( $\rho = -0.55$ , p = 0.019). oFSI correlated significantly with only total organic sediment ( $\rho = -0.55$ , p = 0.016) whilst ToFSI was not significantly correlated with any fine sediment metrics. Notably, total sediment was also not significantly correlated with PSI ( $\rho = -0.54$ , p = 0.018), EPSImixed ( $\rho = -0.58$ , p = 0.008), oFSI ( $\rho = -0.55$ , p = 0.016) and CoFSI ( $\rho = -0.52$ , p = 0.046). The suspended sediment metric, background SSC, was not correlated significantly with any sediment-specific index. The correlation matrix of EQRs (see p. 31 for explanation) for sediment-specific biomonitoring indices can be found in Appendix 3.15.

	PSI													
PSI	1	EPSI	nixed											
EPSI	0.85*	1	EPSII											
EPSImixed	0.87*	0.95*	1	oFSI										
oFSI	0.79*	0.65*	0.74*	1	ToFS		each							
ToFSI	0.06	0.23	0.30	0.02	1	CoFS	l fines r							
CoFSI	0.52*	0.55*	0.66*	0.64*	0.73*	1	Visua	surface	ace					
Visual fines reach	-0.57*	-0.60*	-0.65*	-0.37	-0.45	-0.54*	1	Total	nic surfa	face				
Total surface	-0.53	-0.51	-0.59*	-0.39	-0.41	-0.55*	0.82*	1	Orgar	anic sur	nt			
Organic surface	-0.50	-0.41	-0.50	-0.48	-0.18	-0.51	0.53*	0.65*	1	Inorga	sedime			
Inorganic surface	-0.51	-0.49	-0.56*	-0.36	-0.41	-0.54*	0.82*	0.99*	0.63*	1	Total	organic	<u>.</u>	
Total sediment	-0.45	-0.45	-0.50	-0.35	-0.29	-0.43	0.73*	0.92*	0.56*	0.91*	1	Total	inorgan	SSC
Total organic	-0.54*	-0.50	-0.58*	-0.55*	-0.17	-0.52*	0.62*	0.74*	0.89*	0.73*	0.76*	1	Total	Jround
Total inorganic	-0.44	-0.44	-0.49	-0.33	-0.31	-0.44	0.73*	0.92*	0.55*	0.92*	1.00*	0.75*	1	Backg
Background SSC	-0.28	-0.23	-0.29	-0.25	-0.03	-0.13	0.22	0.23	-0.21	0.23	0.36	0.06	0.34	1

Figure 5.16 – Spearman's rank correlation matrix of observed sediment-specific biomonitoring index scores with different metrics of fine sediment. Asterisks show significant correlation pairs.

No non-specific indices performed better than fine sediment-specific indices with either visual fines, total surface or total sediments (Figure 5.17). However, several indices responded to the organic metrics better than the sediment-specific indices. Organic surface sediment correlated significantly with EPT ( $\rho = -0.56$ , p = 0.024). Total organic sediment correlated significantly with WHPT\_ASPT ( $\rho = -0.59$ , p = 0.008), BMWP\_ASPT ( $\rho = -0.60$ , p = 0.006), and EPT ( $\rho = -0.66$ , p = 0.004). The correlation matrix of EQRs for each biomonitoring index can be found in Appendix 3.15. When considering the relationship between biomonitoring indices, there was a strong correlation between PSI derived indices (Figure 5.18). There was also a strong correlation

between LIFE with PSI ( $\rho$  = 0.86, p <0.001), EPSI ( $\rho$  = 0.74, p <0.001), EPSImixed ( $\rho$ = 0.76, p <0.001) and oFSI ( $\rho$ = 0.72, p <0.001). The strongest pairwise correlation between any of the CoFSI derived indices and a nonspecific index was oFSI and EPT ( $\rho$ = 0.86, p <0.001). There was a strong correlation of FRic with WHPT\_NTAXA ( $\rho$ = 0.87, p <0.001), BMWP ( $\rho$ = 0.72, p <0.001), and BMWP\_NTAXA ( $\rho$ = 0.83, p <0.001). There was no links between the functional indices and sediment-specific indices.



Figure 5.17 - Correlation matrix of observed non-specific biomonitoring indices with different metrics of fine sediment. Asterisks show significant correlation pairs.



Figure 5.18 – Correlation matrix of observed sediment-specific indices and nonspecific indices. Asterisks show significant correlation pairs.

The model results for PSI, EPSI, EPSImixed and CoFSI are presented in Tables 5.17 - 5.20 (model results for all other indices can be found in Appendix 3.16; diagnostic plots for all models can be found in Appendix 3.17). PSI, EPSI and EPSImixed models were relatively well fitted with high adj R<sup>2</sup> values (0.65, 0.68 and 0.72 respectively). The CoFSI model was less well fitted (R<sup>2</sup> = 0.42), conversely the oFSI model had a much better fit (R<sup>2</sup> = 0.64). The antecedent flow metric, Q20pre7d (the flow that was exceeded 20% of the time in the 7 days prior to sampling), was significant for PSI, EPSI and EPSImixed (Table 5.6 - 5.7 for acronym definitions of flow metrics). In each case, this antecedent flow metric had the largest coefficient estimate size and was therefore the most important variable controlling index score. The coefficient estimate was negative which means that the higher the flow in the previous 7 days, the lower the index score (i.e. the indices are indicating higher sediment stress). The antecedent flow metric Q20pre6m was significant for PSI only. Filamentous algae, stream power (both negative coefficient estimates) and coarse bed matrix (positive coefficient estimate) variables were significant for PSI, EPSI and EPSImixed. Detritus was significant with a negative estimate for EPSImixed. It is intuitive that a higher proportion of detritus will result in lower index scores (i.e. higher sediment stress). End group, depth and bedrock were the only three variables significant in the CoFSI model.

Response	Coefficient	Estimate	Std. Error	t value	р
	(Intercept)	20.617	13.484	1.529	0.137
	Q1090DF	-11.560	7.731	-1.495	0.145
PSI	Q20pre6m	-23.460	-23.460 8.948		0.014*
	Q20pre7d	-38.304	10.085	-3.798	0.001*
<b>df</b> 30	Detritus	-3.415	1.802	-1.895	0.068
Adj R <sup>2</sup>	Filamentous algae	-3.747	1.836	-2.040	0.050*
0.653	Altitude	3.309	1.930	1.715	0.097
<b>F</b> 8.026	Slope	6.889	2.346	2.936	0.006*
<b>p</b> <0.001*	Distance from source	-4.742	2.225	-2.131	0.041*
	Coarse bed Matrix	6.234	2.179	2.861	0.008*
	Stream power	-7.797	2.231	-3.494	0.001*
	Season (spring)	4.946	3.542	1.396	0.173

Table 5.17 – Refined linear model results for PSI. Significant effects are marked with an asterisk.

Response	Coefficient	Estimate	Std. Error	t value	р
	(Intercept)	61.536	4.365	14.096	<0.001*
	Q20pre6m	-13.268	7.371	-1.800	0.082
EPSI	Q20pre7d	-31.130	9.439	-3.298	0.002*
	Bedrock	-2.494	1.583	-1.575	0.125
<b>df</b> 31	Detritus	-3.250	1.655	-1.964	0.059
<b>Adj R<sup>2</sup></b> 0.686	Filamentous algae	-4.266	1.702	-2.507	0.018*
<b>F</b> 9.945	Macrophyte	-5.771	1.827	-3.159	0.004*
<b>p</b> <0.001*	Slope	3.690	2.429	1.519	0.139
-	Distance from source	-2.880	1.827	-1.576	0.125
	Coarse bed matrix	6.259	1.911	3.276	0.003*
	Stream power	-6.626	2.099	-3.157	0.004*

Table 5.18 – Refined linear model results for EPSI. Significant effects are marked with an asterisk.

Table 5.19 – Refined linear model results for EPSImixed Significant effects are marked with an asterisk.

Response	Coefficient	Estimate	Std. Error	t value	р
	(Intercept)	56.221	4.547	12.363	<0.001*
EPSImixed df 29	Q20pre6m	-13.715	7.357	-1.864	0.072
	Q20pre7d	-39.412	9.885	-3.987	<0.001*
	End group	-5.145	2.652	-1.940	0.062
	Depth	3.439	2.101	1.637	0.112
	Bedrock	-3.627	1.787	-2.030	0.052
<b>Adj R</b> <sup>2</sup> 0.716	Detritus	-4.437	1.706	-2.601	0.014*
<b>F</b> 9.622	Filamentous algae	-6.109	1.711	-3.570	0.001*
<b>p</b> <0.001*	Macrophyte	-5.833	1.843	-3.165	0.004*
	Altitude	2.658	1.778	1.495	0.146
	Distance from source	-7.035	2.588	-2.718	0.011*
	Coarse bed Matrix	8.356	1.938	4.311	<0.001*
	Stream power	-4.044	1.852	-2.183	0.037*

Response	Coefficient	Estimate	Std. Error	t value	р
CoFSI	(Intercept)	4.576	0.029	159.960	<0.001*
	End group	-0.119	0.030	-3.996	<0.001*
<b>df</b> 36	Depth	-0.099	0.030	-3.268	0.002*
Adj R <sup>2</sup>	Bedrock	-0.078	0.031	-2.535	0.016*
<b>F</b> 6.949 <b>p</b> <0.001*	Detritus	-0.049	0.029	-1.655	0.107
	Filamentous algae	-0.048	0.030	-1.627	0.112

Table 5.20 – Refined linear model results for CoFSI. Significant effects are marked with an asterisk.

It is important to consider the differences in how the indices perform at each site in the context of WFD classification. The EQRs for the sediment-specific indices are plotted in Figure 5.19 and for the non-specific indices in Figure 5.20. In the top left panel plot for each index, the dashed line represents the current good/poor boundary. Although PSI has not been officially adopted, Extence et al. (2017) suggests EQRs between 0.8 – 1.2 are considered normal (meeting prediction). EQRs below 0.8 are falling below predictions (have excessive fine sediment stress) and those above 1.2 are exceeding prediction. There are no boundary classifications for CoFSI. The WHPT ASPT boundary is 0.86 and for LIFE it is 0.94. The sites with the lowest EQRs are highlighted on each graph which vary according to each index. Site 177Autumn consistently falls below the good/poor boundary for PSI, EPSI, EPSImixed and CoFSI but is within the good boundary for WHPT. Considering WHPT is an index for general health, only four sites fall below the 0.86 boundary. However, for the sediment-specific indices, 10 sites fall below the boundary for PSI and seven sites for both EPSI and EPSImixed. WHPT\_ASPT, as a well-developed index which has undergone several optimisations, is often used to compare with the performance sedimentspecific indices. A low NTAXA indicates poor habitat quality whereas low ASPT indicates poor water quality. Seven sites fall below the boundary line for WHPT\_NTAXA indicating more sites with lower habit quality.



Figure 5.19 – O:E plots for PSI O:E (a), by season (b) and RIVPACS endgroup (c); EPSI O:E (d), by season (e) and RIVPACS endgroup (f); EPSImixed O:E (g), by Season (h) and RIVPACS endgroup (i); and CoFSI O:E (j), by season (k) and RIVPACS endgroup (I).


Figure 5.20 – O:E plots for WHPT\_ASPT O:E (a), by season (b) and RIVPACS endgroup (c); WHPT\_NTAXA O:E (d), by season (e) and RIVPACS endgroup (f); and LIFE O:E (g), by season (h) and RIVPACS endgroup (i).

### 5.4.4 Indicator taxa, traits and turnover

TITAN analysis showed nine taxa with a threshold for decreasing across a gradient of visual fines and only one taxon, *Sialis lutaria*, increasing (Figure 5.21a). *Sialis lutaria*, along with Dytiscidae, was also identified as an indicator which increases across the gradient for total organic surface sediment. For total surface and total sediment, all taxa identified by TITAN were indicators with a threshold for decreasing as the value of the metric increases. There were many species in common identified across the different metrics of fine sediment. *Baetis scambus* and *Hydropsyche siltalai* were the only taxa identified for all four metrics. The threshold that elicited change was relatively low for all metrics, but appeared lowest for total organic surface.



Figure 5.21 – Results of taxonomic TITAN analysis for visual fines (%) (a) total surface sediment (g m<sup>-2</sup>) (b), total sediment (g m<sup>-2</sup>) (c) and total organic surface sediment (g m<sup>-2</sup>) (d). Circles indicate change points and whether the taxa show an increase (Z+) or decrease (Z-) in abundance along the gradient. The size of the circles represents the Z-score (i.e. the strength of the response to the fine sediment gradient) and horizontal lines represent 5 – 95% confidence intervals for each taxon.

The TITAN analysis of trait data showed five trait modalities (Aqs.Egg, Dis.AquaticActive, Rif.EggsStatoblasts, Foo.Microphytes, and Fee.Scrapers) (see Table 5.8 for abbreviations) with a threshold beyond which they decrease across a gradient of visual fine sediment (Figure 5.21). Two traits (Rep.ClutchesFree and Foo.Microorganisms) had a threshold indicating they increase in abundance across the gradient. For total surface sediment, five trait modalities (Aqs.Egg, Rep.ClutchesFixed, Foo.Microphytes, Fee.Scraper, and Dis.AquaticActive) showed a threshold indicating they decrease with the total surface sediment metric and four (Dis.AerialPassive, Rep.ClutchesFree, Aqs.Nymph, and Foo.Vertebrates) had a threshold indicating an increase. For total sediment and organic surface sediment, all except one trait (Rep.Ovovivparity for total sediment and Foo.Vertebrates for total organic surface) identified were indicators for decreasing with fine sediment. There were several similarities across all metrics but only Aqs.Egg was consistently identified across all metrics. The threshold that elicited change was relatively low under all sediment metrics, however slightly higher than the levels for taxonomic TITAN analysis.



Figure 5.22 – Results of trait TITAN analysis for visual fines (%) (a) total surface sediment (g m<sup>-2</sup>) (b), total sediment (g m<sup>-2</sup>) (c) and total organic surface sediment (g m<sup>-2</sup>) (d). Circles indicate change points and whether the taxa show an increase (Z+) or decrease (Z-) in abundance along the gradient. The size of the circles represents the Z-score (i.e. the strength of the response to the fine sediment gradient) and horizontal lines represent 5 – 95% confidence intervals for each trait.

The RLQ result indicated that the overall taxa-trait-environment link was marginally significant (p = 0.041). The first axis represents the majority of the variation in the RLQ analysis (Appendix 3.18) and therefore most observations will be focussed on the first (x) axis only. The RLQ plots suggest a separation from higher altitude, steeper sites towards the right of the ordination plots to more lowland, shallower and plant rich sites which are depositing more fine sediment towards the left of the plots (Figure 5.23). This aligns with taxa and trait shifts from more taxa in EPT families towards the right to Chironomidae, Hirudinea and filter or deposit feeders towards the left. However, no significant individual taxa-trait-environmental pairs were significant and the marginal significance of the overall analysis indicate that any observations from the RLQ plots must be treated with caution.



Figure 5.23 – Fourth corner analysis results for taxa (a), sites (b), traits (c) and environmental variables (d).

The results of the GF for the visual fines metric show six species exhibiting a significant turnover in relative abundance as fines increased (Table 5.21 - 5.22). Turnover can be defined as the rate of species replacement and change in abundance between sites (Baselga and Orme 2012). The strongest responding taxon (highest R<sup>2</sup>) was *Proasellus meridianus*. Three traits, Dis.AquaticActive, Fee.DepositFeeder and Fee.Shredder, showed a significant turnover (see Table 5.10 for full names). The strongest relationship was for Fee.Shredders. Six taxa and three traits were identified by the GF for total surface sediment Table 5.21). The strongest responding taxa was *Limnophila* sp and the strongest responding trait was Foo. Microphytes (microphytes as a food type). The only similarity between the visual fines and total surface GF was *Ecdyonurus* sp. Species turnover appears more gradual over the visual fine sediment gradient however most of the turnover occurs in the first 20% of the total surface sediment gradient (Figure 5.24). In both cases, taxonomic turnover reaches the maximum before trait turnover. When comparing the importance of each environmental variable to the overall taxa and trait GFs (i.e. incorporating all sediment metrics and the reduced set of environmental variables), antecedent flow metrics and altitude appeared the most important (Figure 5.25).

Visual fines								
Таха	R²	Traits	R²					
Agapetus sp.	0.009	Dis.AquaticActive	0.049					
Ecdyonurus sp.	0.009	Fee.DepositFeeder	0.029					
Glossiphonia complonata	0.107	Fee.Shredder	0.114					
Helobdella stagnalis	0.026							
Proasellus meridianus	0.212							
Sphaereum corneum	0.068							

Table 5.21 – Significant taxa and traits for from the gradient forest analysis for visual fines.

Table 5.22 – Significant taxa and traits for from the gradient forest analysis for	
total surface sediment	

Total surface								
Таха	R <sup>2</sup>	Traits	R <sup>2</sup>					
Ecdyonurus sp.	0.148	Foo.Microphytes	0.107					
Empididae	0.012	Foo.Microinvs	0.046					
Esolus parallelepipedus	0.104	Fee.Scraper	0.052					
Hydropsyche siltalai	0.063							
Lepidostoma hirtum	0.145							
<i>Limnophila</i> sp.	0.284							



Figure 5.24 – Standardised turnover from gradient forest analysis for visual fines (a) and total surface sediment (b). Each individual measurement of total surface sediment is shown in the rug on the x axis.



Figure 5.25 – Gradient forest importance of environmental variables for taxa (a) and trait (b) turnover.

### 5.5 Discussion

The aim of this chapter was to: compare existing methods of measuring fine sediment; test the performance of sediment-specific biomonitoring indices; compare the performance of other non-specific indices; determine which environmental variables affect index performance, and assess indicator taxa, trait and trait-environment relationships using a range of statistical methods. This was achieved through a field sampling regime which sampled 21 sites over two seasons collecting macroinvertebrates, environmental variables (including hydrological data), and quantified different metrics of fine sediment. This is the first study to independently test existing sediment-specific biomonitoring tools in the UK. By interpreting the results of this study, recommendations for future academic research and application of monitoring practices can be made.

### 5.5.1 Comparing methods of measuring fine sediment

Methods for quantifying sediment in rivers are best classified depending on whether they quantify suspended or deposited sediment. In Chapter 2 (Section 2.4.1.1 and 2.4.1.2), various methods of measuring suspended and deposited sediment were described. In this study, the two methods of measuring deposited fine sediment were the visual estimate method (a semi-quantitative rapid assessment) and the fully quantitative resuspension method (Lambert and Walling 1988, Duerdoth et al. 2015). These methods were chosen as they were used in the development of EPSI and CoFSI respectively.

This section of the study builds on work by Conroy et al. (2016b) who compared various methods of measuring fine sediment in laboratory-based mesocosms and recommended further comparisons under field conditions. The present study showed a strong and significant correlation between reach scale visual estimates and total surface sediment. The results of the present study support that of Zweig and Rabení (2001) and Glendell et al. (2014) who found that the measure of embeddedness and visual estimates were highly correlated with one another. Hubler et al. (2016) showed correlations of between 0.49-0.58 which is lower than the present study. However fine sediment was defined by Hubler et al. (2016) as particles <0.06 mm in diameter (which is smaller than the size defined within the present study) potentially indicating that visual observations are insufficient at identifying particles at this size. Duerdoth et al. (2015), showed inter-operator variability was a significant influence accounting for up to 40% of the total variance of visual estimates. Within the present study, inter-operator variability was eliminated (as the same operator assessed fine sediment at each site) which could account for the stronger correlations

between the semi-quantitative and fully quantitative metrics. The correlation between visual estimates and total surface sediment was stronger when the visual estimates were taken at the patch scale. This is expected, considering the patch scale estimates were taken of the undisturbed area of bed surface within the resuspension cylinder prior to agitation. This is perhaps confounded and a more appropriate comparison may be to examine a set of random patches within the sampled reach. However, it provides additional support for the visual estimates, not least because of the closer relationship between the fully quantitative and semi-quantitative measures at the patch scale, but also because the accuracy of visual estimates is not drastically reduced at the reach scale.

When comparing the relationship between total surface sediment and visual estimates by season, the correlation was stronger in spring than in autumn. The weaker fit in autumn could have been a result of leaf litter and other detritus obscuring views of fine sediment and resulting in underestimates. Or alternatively, high organic content on the riverbed from leaf litter breakdown could lead to overestimations. However, a linear modelling approach showed season did not significantly affect the overall relationship between visual estimates and total surface sediment. The weaker link between the organic surface and the total organic content with all other metrics of fine sediment suggests that the organic content is relatively independent of the total sediment content and is likely dependent on other factors which influence the supply and breakdown of organic matter.

Visual estimates correlated well with the total estimates. The subsurface agitation incorporates both the surface drape and the sediment within the top 100 mm of the gravel bed. Criticism of EPSI has focused on the lack of representativeness of the visual estimates which only estimate the surface drape which may not necessarily be associated with the subsurface sediment. Subsurface sediment can be transported laterally in the subsurface of gravel bed rivers, and its retention and accumulation is an important part of the sediment transport system (Harper et al. 2017). Studies deploying sediment traps in situ within the river bed have shown lateral sediment movement to contribute between 20-46% of total surface and subsurface sediment mass (Carling 1984, Sear 1996, Mathers and Wood 2016). Additionally, rivers dominated by vertical sediment ingress can lead to the formation of seals or clogs blocking further sediment movement by vertical exchange (Frostick, Lucas, and Reid 1984). Most macroinvertebrates live in the upper layer of sediment in gravel beds. Therefore, the surface sediment layer is potentially the most ecologically important metric of fine sediment that should be considered. However, the present study has shown the visual estimates are representative of the subsurface sediment.

Challenges were encountered when using the resuspension method. Achieving a seal around the cylinder, in order to prevent sediment winnowing, was particularly challenging in reaches dominated by coarse substrates. As described in Section 5.3.2, two resuspension samples were taken from erosional areas and two from depositional areas in order to sample the two extremes of fine sediment retention in river channels (Collins and Walling 2007a, 2007b, Duerdoth et al. 2015). In some reaches there was no clear distinction between erosional and depositional areas. In these instances, the process of identifying appropriate areas to sample was particularly subjective. Additionally, there could potentially be broader problems with the representativeness of sampling in these two extremes.

When modelling each sediment metric as a function of environmental variables, flow metrics, particularly antecedent metrics, appeared most important in predicting the deposited sediment metrics. Flow is intrinsically linked to sediment supply, transport and retention in rivers (Van Rijn 1993). High discharges have sufficient stream power to carry larger and greater amounts of fine sediment in suspension. This results in deposited sediments being cleared from the river bed, and suspended sediment increasing, providing stream power is maintained. Continual or uncharacteristically low flows can result in increased deposition of fine sediment on river beds. This aligns with the results from the present study. The antecedent flow metrics Q50preSum (significant for visual fines and total surface) and Q50preWin (significant for visual fines and total surface) all

had positive coefficient-estimates (i.e. as they increase, the quantity of fine sediment also increases) (see Table 5.6 and 5.7 for hydrological metric abbreviations). This is intuitive for deposited sediment metrics although no antecedent or flow regime variables were significant for total sediment. Erosional flow (proportion of erosional flow types within sampling reach) was significant for total sediment, perhaps indicating that site specific hydraulic conditions are more important than overall flow patterns in influencing subsurface infiltration (see Figure 2.1). The higher antecedent flow variable, Q20pre6m was significant for background SSC with a negative coefficient, perhaps indicating a link with the effects high flows have on sediment supply in the catchment (Lawler et al. 2006). The variance explained by the linear model for SSC was particularly low compared to the deposited metrics. Thus, unsurprisingly, suspended sediment is poorly explained by the same set of environmental variables as deposited sediment. Despite large variations in deposited sediment metrics between sites, there was low variation of SSC. This is also supported by SSC contributing a low proportion of the overall variability of the PCA (Figure 5.8). This is because sampling was only carried out during low flow (high and spate flows were avoided) and therefore little variation in SSC was captured.

Season was a significant predictor where it was included as a fixed effect (total sediment and background SSC). Season was also significant for the organic metrics, further reflecting the variation in organic matter supply seasonally. Most studies to date which compare the semi-quantitative estimates with the fully quantitative resuspension method only sample a single season, missing this ecologically relevant variation. Width was a significant predictor for both the deposited metrics and background SSC, with negative coefficient estimates (i.e. as width increases, the estimates of fine sediment will decrease). Width is closely linked to both discharge and velocity and therefore the effect of width could be a proxy for these effects. Given that width is a significant predictor, this could imply that small streams are most vulnerable to fine sediment accumulation and could indicate where resources are best allocated in catchment management projects. Notably, stream power was not included in

the reduced models. This unexpected result could be because the effects are captured by other variables (e.g. flow variables). The coarse bed matrix was a significant predictor of visual estimates, total surface sediment and total sediments. The calculation of the coarse bed matrix is not completely circular with the percentage of fine sediment (as it does not include the percentage of gravel present), however this result is predictable. Additionally, flow patterns around coarse substrates can create hydrodynamic conditions which resuspend deposited sediments (Buffin-Bélanger and Roy 1998).

# 5.5.2 Comparing the performance of sediment-specific biomonitoring indices

There were several hypotheses relating to the performance of sediment-specific indices. It was hypothesized that EPSI and CoFSI would show a stronger relationship than PSI with metrics of deposited fine sediment. The reason behind this is because both CoFSI and EPSI use empirical (both sediment and ecological community) data to calibrate the index, whereas PSI is based purely on sensitivity weightings assigned using expert knowledge. It was also hypothesized that EPSI would show a stronger relationship with visual estimates of fine sediment and, conversely, that CoFSI would show a stronger relationship with quantitative estimates of deposited fine sediment (as these were the metrics used to calibrate each index respectively). There was evidence to partially support these hypotheses.

All PSI derived indices correlated significantly with visual fines, with correlation coefficient size following the pattern PSI < EPSI < EPSImixed reflecting the improvement of each subsequent iteration of the index. The stronger performance of EPSImixed supports evidence in Chapter 3 (Section 3.4.2) which showed that responses are more likely to be detected when taxa are described at a coarser identification level than species. The EPSImixed index contains more mixed taxa level (i.e. species, genus and family) scoring than EPSI. PSI and EPSImixed correlated well with total surface sediment. Therefore, despite calibration with only a semi-quantitative measure, this index still performs well with the fully quantitative method. This further supports the

evidence that visual estimates (i.e. surface drape) are a reliable proxy for quantitative measures of total surface sediment (Glendell et al. 2014, Conroy et al. 2016b). Total sediments were not significantly correlated with any sedimentspecific index or non-specific index. However, the total organic sediment was significantly correlated with PSI, EPSImixed, oFSI and CoFSI.

CoFSI was significantly correlated with both total surface and visual fines. It performed better than PSI when correlated with total surface sediment, however it did not perform as well as EPSImixed. Furthermore, it did not perform as well as any of the PSI derived indices with visual fines. The oFSI constituent of CoFSI also correlated poorly with all metrics, with the exception of total organic sediment. The oFSI component was derived from the organic mass in erosional areas and it would have been expected to show a strong association with organic surface and total organic measures. ToFSI was not significantly correlated with any metric of fine sediment. The ToFSI component relates to the total fine sediment mass in the surface drape of depositional areas. The relatively poor performance of CoFSI is in stark contrast to its performance in previous studies (see Table 5.2; Murphy et al. 2015; Turley et al. 2016). During the development of CoFSI, the taxa with the lowest 10% abundance were removed. This potentially eliminates a large number of taxa conforming to Kselected life strategies (e.g. large body size, longer life expectancies, produce fewer offspring). Taxa of this type are likely to be less tolerant of stress and disturbance and therefore more sensitive to fine sediment (Weinbauer and Hofle 1998). Thus, their inclusion in a stressor-specific index is imperative. The CoFSI removal process eliminated 208 taxa, leaving the index with only 105 scoring taxa. Furthermore, the final calculation is based on presence/absence only. The method used in constructing CoFSI (described in Section 5.1.1) is relatively common in the development of biomonitoring indices (Birk et al. 2012). However, it lacks a mechanistic basis (Wilkes et al. 2017). Collectively, the low number of scoring taxa, the removal of potentially sensitive species and based purely on presence/absence combines to create a poorer performing index.

It was also hypothesized that all sediment-specific indices will show a stronger relationship with fine sediment than non-specific indices. There is evidence to fully support this hypothesis. There were no significant correlations between non-specific indices with total surface sediment or visual fines. Observing the correlations between each of the indices (both sediment-specific and nonspecific) can help understand the mechanics of each index. Flow is inherently linked to sediment transport and retention. Species which are sensitive to excessive fine sediment are also sensitive to low flow conditions (Extence, Balbi, and Chadd 1999, Extence et al. 2013). This is shown by the strong correlation between LIFE and all sediment-specific indices. All PSI derived indices are closely related. The correlation of PSI with LIFE was stronger than with EPSI and EPSImixed. Similarly, to PSI, the LIFE index was also developed based on expert knowledge only. Taxa and traits which possess sensitivities to low flow (i.e. score as sensitive on the LIFE index) will most likely align with the taxa and traits which are sensitive to fine sediment. It is difficult to separate the specific abiotic conditions that organisms are responding to, e.g. higher velocity habitats which also accumulate less fine sediment and the resultant oxygen conditions within the water. It is likely a combination of both flow and sediment. This has been recognised by Turley et al. (2017), however when EPSImixed was tested over a range of stream powers, the relationship weakened as stream power increased. The lower correlation values between EPSI and EPSImixed with LIFE, compared to PSI with LIFE, are likely a result of the optimisation specifically with empirical data of fine sediment. This demonstrates an improvement in the specificity of the index (Rosenberg and Resh 1993).

The oFSI constituent of CoFSI had close associations with non-specific indices. As the oFSI is derived from the mass of organic sediment in erosional areas, it would be expected that this index would be closely linked to BMWP and WHPT. Despite undergoing several rounds of optimisation, and often used as indices for general ecological health, BMWP and WHPT were developed originally based on organic pollution sensitivity (Armitage et al. 1983, Paisley, Trigg, and Walley 2014). High organic content in fine sediments can cause chemical changes in the benthic zone and reduce oxygen availability for aquatic

organisms (Von Bertrab et al. 2013). When modelling individual taxa scores for the oFSI index with their corresponding trait scores, Wilkes et al. (2017) showed oFSI to be strongly related to traits describing respiration. This corroborates with existing knowledge on the effects of organic stress on aquatic organisms. The results of this study support that the oFSI element of the CoFSI index is detecting a response to organic stress. However, other non-specific indices also correlated well with organic metrics. The EPT index was significantly correlated with organic surface sediments and WHPT\_ASPT, BWMP\_ASPT and EPT correlated significantly with total organic sediment. There are two theories to explain these interactions. Considering the significant correlation of total organic sediment with PSI, EPSImixed, oFSI and CoFSI, this could suggest that there is a common adaptation to both stressors. Alternatively, the pressure from fine sediment could be synonymous with organic pressure which is predominantly linked to the increased sediment oxygen demand (see Chapter 2 Section 2.3.2). Given these two theories, the effects of organic pressure are likely indistinguishable from those of fine sediment. Therefore, the ability of any sediment-specific index to detect fine sediment pressure demonstrates they are well developed and appropriate for use.

The correlation values between fine sediment and each of the indices in this study were all lower than those found in the original studies from each of the index developments. This study limited the river type to lowland gravel bed rivers, whereas EPSI and CoFSI calibration studies looked at all river types in England and Wales. Additionally, the focus of both EPSI and CoFSI research was sediment from agricultural sources (determined through GIS and site selection processes) whereas in the case of this study, all sediment stress was considered. Despite visual estimates of fine sediment ranging from between 0 - 100% and quantitative metrics of fine sediment ranging across several orders of magnitude, most of the sites scored towards the higher end of the sediment-specific index ranges (i.e. indicating low fine sediment pollution). The suspended sediment metric, SSC, was not associated with any indices which is in contrast to the moderate correlation of PSI with suspended solids (mg l<sup>-1</sup>) as shown in Turley et al. (2014). However, the suspended solids metric from Turley

et al. (2014) were derived from the mean of a minimum of 12 SSC samples taken over at least a one-year period at each site, perhaps indicating a better estimate of background SSC as opposed to the two samples (one for each season) taken in the present study.

When modelling the predictors of index performance, significant coefficientestimates were similar between PSI, EPSI and EPSImixed. Antecedent flows appeared to be the most important determinants of index scores. The antecedent flow metric Q20pre7d was significant for all sediment-specific indices, and was also the predictor with the largest coefficient-estimate size (i.e. causes the largest variation in index scores). This antecedent metric quantifies relatively high flow rates in the previous 7 days before sampling took place. The coefficient-estimates were all negative, this means that as the Q20pre7d increases (i.e. higher flows), the index score decreases (i.e. higher deposited sediment stress). This is counterintuitive because recent high flows should have a flushing effect, removing fine sediment from the bed. A biological explanation could be that the recent antecedent high flows have stimulated insect dispersal through drift or reduced abundance through scour of individual organisms (as a result of increased velocities or potentially suspension of fine sediment) (Mackay 1992, Svendsen, Quinn, and Kolbe 2004). A decrease in the index scores could have been a result of a reduction in species which also possess sensitivities to sediment (i.e. score as sensitive). A geomorphological explanation could be that high flows in the preceding period could deliver more sediment to the reach, and the reduction in index score is reflecting an actual effect of fine sediment increase. Similarly, the antecedent flow index Q20pre6m was significant for PSI. Relatively high discharges could have affected macroinvertebrate recruitment at a key time in the macroinvertebrate reproductive cycle. The significance of these flow metrics in the model could be a result of flow effects on macroinvertebrates as opposed to a direct link with fine sediment (Dunbar et al. 2010).

River longitudinal slope was a significant predictor of PSI with positive coefficient-estimates. River longitudinal slope is closely linked with flow velocity and stream power. Stream power was also significant, albeit with a negative

coefficient-estimate. The significance of this in the model could be due to the association with flow sensitivities as described above rather than the direct impacts of fine sediment. As the variability of slope would have been low across all sites, the effect of stream power will likely be dominated by the effects of discharge and, therefore, its effects were more closely captured by the flow metrics included in the models. Rivers with high stream power are less likely to be transport limited (Naden et al. 2016) and are less likely to accumulate fine sediment.

Similar to the sediment metrics (Section 5.5.1), the coarse bed matrix was significant for most of the sediment-specific indices (PSI, EPSI and EPSImixed). The coefficient estimate was positive, which means that the higher the proportion of coarse substrates, the higher the index score (lower sediment stress). The presence of coarse substrates provides a heterogeneous habitat for macroinvertebrate communities. Additionally, flow patterns around coarse substrates can create hydrodynamic conditions which resuspend deposited sediments (Buffin-Bélanger and Roy 1998). Filamentous algae was significant for PSI, EPSI and EPSImixed with negative coefficient estimates. Filamentous algae has the potential to bind and retain deposited fine sediment on the bed surface (Lee, Hur, and Toorman 2017, Wilkes et al. 2019). Macrophytes were significant for EPSImixed, also with a negative coefficient. In-channel or marginal macrophytes can slow near-bed velocities and increase sediment deposition (Sand-jensen 1998, Jones et al. 2012a). Both the presence of filamentous algae and macrophytes have the potential to increase fine sediment stress resulting in lower index scores.

The CoFSI index was poorly predicted by the same set of environmental variables as the PSI derived indices. In contrast, the environmental variables retained in the oFSI and ToFSI models resembled those of the PSI derived indices. The CoFSI score is directly dependent on the individual oFSI and ToFSI values (Appendix 3.1). Season was not included in the optimal models of any of the PSI derived indices or for CoFSI (however it was included for oFSI and ToFSI). Currently, standard national monitoring practice is to sample macroinvertebrate communities during spring and autumn and an average

score is provided for environmental health assessments. If season does significantly affect the response of an index, then providing an average score across both seasons may lead to the misidentification of stream reaches affected by fine sediment pressure.

When considering the sites which were 'failing' under current WFD quality boundaries, there were similarities across PSI derived indices. When comparing with WHPT\_ASPT and WHPT\_NTAXA, many more sites fell below the recommended target for PSI, EPSI and EPSImixed than the non-specific indices. This potentially shows a specificity of these sediment-specific indices to detect poor environmental conditions relating to fine sediment that the indices used for general health are missing. As there are no recommended boundaries for the CoFSI index, it is difficult to determine which sites are failing and therefore compare with other indices.

## 5.5.3 Taxa-trait-environment relationships

Most of the taxa identified as indicator species by the TITAN analysis were from the EPT families. TITAN aims to detect congruence in taxon-specific changes of abundance and occurrence along an environmental gradient. Many of the same taxa were selected as sensitive across visual fines and total surface sediment. Total sediment had only five taxa and five traits identified as indicators. This could reflect that the total sediment (which includes the sediment in the top 100 mm of gravel) is less ecologically relevant than total surface alone. Sialis lutaria and Dytiscidae were the only taxa to show a threshold of increasing abundance across both total surface and visual fines. Simuliidae was one of the taxa to show a threshold for declining with increasing visual fine sediment. Simuliidae is generally considered tolerant of poorer environmental conditions, and scores low on the WHPT index system. Whereas most EPT taxa exhibited change points below 40%, for Simullidae the threshold point was around 50% and could therefore be considered relatively tolerant. When sediment surface cover is low, this species is likely to persist. However, Simuliidae is a filter feeder and also requires firm stable substrates for attachment (Harding and Colbo 1981).

Therefore, when deposited sediment continues to increase, there appears to be a change point at which it becomes sensitive.

Overall, the TITAN analysis did not identify a large number of taxa as indicators of fine sediment. Recently, Gieswein, Hering, and Lorenz (2019) used TITAN to identify 95 indicator taxa of fine sediment using macroinvertebrate data from 73 stream sites in Western Germany. The higher number of indicator taxa identified could be partly due to the larger number of study sites included in the analysis. However, it appears that when carrying out the TITAN analysis, a lower threshold for determining an indicator was used ( $\geq$ 0.7 as opposed to  $\geq$ 0.95 as recommended by the package developers used for the present study). Therefore, these indicator taxa may be less reliable than those identified from the present study where a more stringent threshold was used in the analysis.

In Chapter 3, one of the most unequivocal trait-environment relationships detected was the negative response of shredders to fine sediment. It was therefore hypothesized that shredders will be sensitive to fine sediment in the present study. There is evidence to partially support this hypothesis. Shredders were not identified by the TITAN analysis. However, the gradient forest analysis showed that shredders were the most significant trait modality for visual fines. This is likely because the TITAN analysis attempts to locate thresholds, whereas gradient forest analysis considers any change in abundance along the gradient (rather than a specific break point). The results of both these analyses can be seen as complementary and therefore the lack of detection in one test does not preclude that of the other. This suggests the shredder response is more complex. The mechanisms behind shredder sensitivity are thought to be associated with burial of leaf litter and a reduction in its quality through inhibition of fungal growth (Couceiro et al. 2010b, Doretto et al. 2016). The results from Chapter 3 also showed burrowers were more likely to exhibit tolerance to fine sediment. The results of this particular study show no such evidence. However, the relationship with burrowers is also complicated. Burrowers can either be sensitive or tolerant depending on what they burrow in to e.g. fine sediment of Caenidae compared to coarser substrates of Ephemeridae (Wilkes et al. 2017).

The RLQ analysis showed a marginally significant (p = 0.041) overall link between traits and environmental variables whilst no individual taxa-traitenvironmental pairs were significant. This is in contrast to Murphy et al. (2017) which found a number of significant relationships. In the present study, the full set of reduced environmental variables were used in the RLQ, compared to only sediment variables in Murphy et al. (2017). However, there are similarities between the traits identified by Murphy et al. (2017) and those identified through the TITAN analysis in the present study (Table 5.23). This is because the TITAN analysis only includes a single fine sediment metric as opposed to incorporating other environment variables. Most of the traits identified by RLQ in Murphy et al. (2017) and in the trait TITAN analysis in the present study may be closely linked to taxonomy and commonly connected with trait syndromes (Verberk, van Noordwijk, and Hildrew 2013). The majority of the traits identified align with those possessed by particular taxonomic groups (e.g. aerial active dispersal and insects). Therefore, incorporating traits in this way does not necessarily improve our mechanistic understanding as it is simply an artefact of detecting taxonomic variation. Traits which would improve mechanistic understanding and not closely associated with taxonomy include body size and diet.

Trait identified by		Т	GF			
Murphy et al.	Visual	Total	Total	Organic	Visual	Total
(2017)	fines	surface	sediment	surface	fines	surface
Aqs.Adult	Х	х	Х	Х	х	х
Rep.Ovoviviparity	Х	х	$\checkmark$	Х	х	х
Dis.AquaticActive	$\checkmark$	$\checkmark$	Х	Х	$\checkmark$	х
Dis.AerialActive	Х	Х	$\checkmark$	$\checkmark$	х	Х
Mod.Crawler	Х	Х	X	Х	х	Х
Rif.EggsStatoblasts	$\checkmark$	Х	$\checkmark$	$\checkmark$	х	Х

Table 5.23 – Similarities between the trait modalities identified in Murphy et al. (2017) from RLQ, with those identified in the present study through TITAN and Gradient Forest (GF) analysis (see Table 5.10 for trait abbreviations).

For both metrics of fine sediment tested using GF, taxonomic turnover (species replacement) plateaued before trait turnover (trait replacement). Most of the taxonomic turnover across the visual fine sediment gradient occurred within the first 50%. For total surface sediment, most of the turnover occurred within the first 20% of the gradient. In the case of total surface sediment, this could be because most of the data were confined to the first 20%. There are two possible interpretations of this result. Most significantly, the impacts of additional fine sediment cause most severe changes to ecological communities when the absolute sediment volume or mass is low. This is significant for managers who need to prioritise limited funding to prevent the worst potential effects. Alternatively, at low sediment stress, the environment is more varied and biotic interactions dominate and therefore there is higher taxonomic and trait turnover. As sediment increases along the gradient, sediment stress will dominate over other environmental variables and turnover will decrease as only tolerant taxa are able to persist. When a range of environmental variables were incorporated into the gradient forest, the results showed that fine sediment metrics had a moderate importance in driving taxonomic and trait turnover (Figure 5.24). This suggests that the most plausible explanation is a mix of these two interpretations. Similar responses to sediment stress have been documented in mesocosm studies (Brown et al. 2019). This is also supported by the TITAN analysis which showed that most of the sensitive taxa exhibited change thresholds below 50%. When considering the importance of sediment metrics in determining the overall taxonomic and trait turnover (i.e. when all environmental variables are considered), sediment metrics show a lower importance than flow (antecedent and flow regime) metrics and altitude. Altitude is generally considered to be a correlate of other variables (e.g. pH, temperature etc), rather than a direct determinant of community structure. The importance of altitude could therefore be reflecting natural regional variations in taxonomic and trait structure. Considering the relatively low importance of sediment metrics in determining taxonomic turnover, this further underlines the success of sediment-specific indices, particularly EPSImixed, to detect fine sediment pressures in the face of variation in other aspects of the environment.

This study sought to test the performance of general and sediment-specific indices and understand which environmental variables affect their performance by measuring a broad selection of abiotic variables. However, there are limitations to this study. Whilst numerous flow and sediment variables were measured and included in the analysis, this study did not incorporate the effect of water quality (e.g. dissolved oxygen, temperature, pH) or sediment quality (e.g. sediment size distribution, presence of sediment associated contaminants). Both of these factors could have significant impacts on macroinvertebrate community structure and therefore affect biological index performance. The purpose of the site filtering process was to remove any sites with poor water quality status which could confound results. The limitations of excluding additional water and sediment quality variables are recognised but the potential impacts of this are minimised by a robust approach to site selection. Additionally, many biotic variables are highly correlated with one another. Whilst the methodology was rigorous in removing collinearity from any modelling analyses, this raises the potential difficulties in isolating cause and effect of fine sediment effects on macroinvertebrates. Therefore, building on conclusions made in Chapter 3 and 4, further research on the mechanisms behind macroinvertebrate responses to fine sediment are required.

### 5.6 Conclusion

The current direction in freshwater monitoring in the UK is to drive forward sediment-specific biomonitoring indices. The results presented in this chapter represent the first full independent assessment of sediment-specific indices developed for use in the UK as well as providing new insights into macroinvertebrate responses to fine sediment. The results showed a strong correlation between PSI derived indices and CoFSI with different metrics of fine sediment. ToFSI performed poorly with all metrics of fine sediment whilst oFSI, the component of CoFSI which is associated to organic stress, was significantly related to total organic sediment. When modelled with other environmental variables, all sediment sensitive indices, with the exception of CoFSI, were highly related to flow variables. Despite large variations in sediment quantity

between sites, antecedent flow seemed to be the overall driving force of index scores. Comparisons of different metrics of fine sediment concluded that visual estimates of fine sediment are a robust proxy for quantitative measures of fine sediment. Building on evidence from Chapter 3, the results of this study show that shredders are sensitive to fine sediment. Functional indices, FDis and FRic, performed poorly and further development of traits-based indices and trait databases is recommended. Taxonomic and trait-based turnover was greatest at low sediment pressures imploring land managers to prioritise the protection to limit the worst potential effects. In summary, the combined results point towards effectiveness of sediment-specific indices, particularly EPSImixed, to detect fine sediment pressures given the close association with organic stress and other environmental variables determining community composition at any given site. It is recommended that this index should be incorporated into monitoring assessment of fine sediment in the UK.

# **Chapter 6 – Conclusions**

# 6.1 Introduction

Fine sediment is considered a significant pressure in aquatic environments globally, and one of the leading causes for failure to meet 'good' ecological status under the WFD (Collins et al. 2011, Mathers et al. 2017). Considering the urgent need to protect freshwater environments from further degradation it is important that fine sediment pressures can be monitored in order to implement appropriate management methods. Freshwater biomonitoring, developed over a century ago, has been integrated into statutory monitoring and assessment since the inception of the WFD (Clarke and Davy-Bowker 2014). More recently, there has been a drive to develop fine sediment-specific biomonitoring indices which are optimised to indicate this particular stressor (Extence et al. 2013, Turley et al. 2015, Murphy et al. 2015). It is important that any biotic index is thoroughly assessed before incorporation into national monitoring frameworks (Birk et al. 2012). Disentangling the multifarious responses of aquatic biota to fine sediment are crucial to developing effective sediment-specific biomonitoring tools. Given this context, the overall aim of this thesis was to quantify the responses of macroinvertebrates to gradients of fine sediment pollution. This overarching aim was subdivided into three smaller aims which were achieved through a number of objectives (see Chapter 2 Section 1.2).

The objectives were accomplished using several different research methodologies; a narrative review, a systematic review, a lab-based study, and a field sampling regime. The following sections will present the fulfilment of each aim through each individual chapter, the key findings, and the implications of the results for both monitoring and academic fields.

### 6.2 Fulfilment of thesis aims and objectives

# 6.2.1 Aim 1 - Identify the main causal mechanisms involved in macroinvertebrate responses to fine sediment

Quantifying the response of macroinvertebrates to gradients of fine sediment is fundamentally multidisciplinary. Considering the research context which was outlined in Chapter 1, Chapter 2 synthesised the knowledge relevant to this aim across the sub-disciplines of geomorphology, hydrology, and ecology. Chapter 2 discussed the key components of the sediment system; sources (delivery), transportation, and deposition. This is crucial to understanding the ecological impacts of fine sediment which are a ultimately a function of its source, quantity, timing of delivery, and retention (Murphy et al. 2015). Chapter 2 also provided a description of the multifarious ways in which fine sediment can affect macroinvertebrates. Suspended sediment (SSC) can affect macroinvertebrates indirectly by increasing turbidity. Increased turbidity reduces light available for photosynthesis by primary producers. The reduced production causes a cascade of effects on the entire trophic system (Nieuwenhuyse and LaPerriere 1986, Klco 2008, Izagirre et al. 2009, Aspray et al. 2017). Increased turbidity also affects predators who rely on visual searching. This predominantly affects top predators, thereby changing top down controls on the trophic system (Breitburg 1988, Boubée et al. 1997, Shoup and Wahl 2009). An increase in fine sediment deposition can alter the supply of nutrients to the gravel bed, resulting in an increase in sediment oxygen demand (Bjornn and Reiser 1991, Sear et al. 2017). Sediment can directly smother the benthos favouring organisms who are able to excavate themselves (Wood et al. 2005, Conroy et al. 2018). Settling and infiltration of fine sediment by the process of colmation clogs the spaces between gravels reducing interstitial water flow critical for the exchange of gases in these pore spaces (Wharton, Mohajeri, and Righetti 2017), thereby restricting the supply of oxygen to benthic organisms and the removal of excreta favouring organisms which are tolerant of these harsh conditions (Owens et al. 2005, Hinchey et al. 2006).

Whilst these are the general mechanisms by which fine sediment can affect biota, the huge functional diversity (FD) of macroinvertebrates makes their response to environmental stressors complex. Chapter 2 provided a valuable contribution to knowledge as it is the first time that the responses of macroinvertebrates to fine sediment have been integrated with the discussion of traditional physical methods of measuring fine sediment, and questions of 'why?' and 'how?' biomonitoring is significant in monitoring the effects of fine sediment. Often reviews only focus on a singular element such as ecological responses (Jones et al. 2012b), bottom-up controls of macroinvertebrates (Wilkes et al. 2019) or fine sediment monitoring (Bilotta and Brazier 2008). It is important that knowledge from each discipline is combined in order to provide a holistic perspective on the problem. However, qualitative reviews of this kind are subject to many biases that can influence their outcomes and conclusions (Møller and Jennions 2001). In spite of this limitation, the narrative formed in Chapter 2 helped to inform the structure of a more systematic style review which was conducted in Chapter 3.

It is clear that there are many ways to describe macroinvertebrate response, e.g. abundance, richness, diversity indices, trait-based responses, behavioural responses, biotic indices etc. However, the evidence reviewed in Chapter 2 was sometimes gathered from confounded research conducted in ways which made direct comparisons difficult. Therefore, a weight of evidence approach to review the responses of macroinvertebrates to fine sediment was appropriate. The aims of Chapter 3 were to quantify the breadth of evidence, classify the types of responses described, assess the weight of evidence for macroinvertebrate responses to fine sediment, and identify any knowledge gaps. Full systematic reviews are labour intensive projects beyond the scope of this thesis. By combining methods from Systematic Mapping (SM) and Rapid Evidence Assessments (REAs) the aims could still be achieved. The results of this chapter showed that there were some consistent responses to fine sediment; shredders were sensitive and burrowers were tolerant to fine sediment. However, there was not enough data to analyse some relationships. There was a global imbalance of fine sediment research and an overall decline of research

outputs when standardised against a more general environmental science term. Chapter 3 provides the first review of its kind in the field of fine sediment research and offers a substantial contribution to knowledge through both the evidence mapping exercise and the evidence assessment. Its conclusions supported the generation of hypotheses for subsequent chapters.

Chapter 4 took a reductionist approach to build on the mechanistic understanding of how fine sediment affects macroinvertebrates. Chapter 2 identified 'abrasion' and 'clogging' as potential effects of suspended fine sediment. However, the evidence for these mechanisms appeared to be weak and indirect. Some individual behavioural responses which have contributed to this theory include resource shifts, reduced feeding rates and active drift (Voelz and Ward 1992, Runde and Hellenthal 2000, Larsen and Ormerod 2010). Many studies which quantify macroinvertebrate responses to fine sediment, and included as part of the review in Chapter 3, often suggested possible mechanisms in a speculative way, but urged research focussed on understanding these responses (e.g. Connolly and Pearson 2007; Cover et al. 2008; Buendia et al. 2013; Culp et al. 2013; Conroy et al. 2016). Mechanisms can be more obvious from experimental studies. Identifying this knowledge gap provided an opportunity to explore the mechanisms behind behavioural responses associated with abrasion and clogging from fine sediment. In a controlled flume experiment, using insect cadavers of varying gill type, the effects of SSC and velocity on gill tissue were studied. Scanning Electron Microscopy images were used to quantitatively analyse the effects on gill tissue. The results showed that potential gill damage in the form of coverage of the gill surface by fine sediment varied by gill type (i.e. species). For *E. danica*, a relatively tolerant burrower, gill coverage did not vary across any treatment. Coverage on *H. siltalai* gill surfaces increased significantly between low and high SSC but only at the higher flow velocity. Finally, for *E. venosus*, the most sensitive species according to sediment-specific indices, SSC influenced gill coverage but velocity had no significant effect. Additionally, there was no evidence of abrasion in the form of scrapes and scratches from the angularity of particles on the gill tissue for any species, raising questions about previous

reviews that have cited this as a key impact of fine sediment on invertebrates (Wood and Armitage 1997, Jones et al. 2012b). This study is the first of its kind to undertake research to understand potential direct, physical effects such as abrasion and clogging and also to apply this novel methodological approach to understand the mechanisms of effect of fine sediment. Whilst this study was not exhaustive, it demonstrates a proof of concept for future work. For example, specimens could be analysed directly from rivers with different levels of sediment pollution.

Although a more holistic approach was taken in Chapter 5, its results also contributed to identifying the main causal mechanisms involved in macroinvertebrate responses to fine sediment by using a variety of statistical methods. The taxonomic and trait-based responses of macroinvertebrates were studied using data from 21 lowland gravel sites sampled over two seasons. The results of Threshold Indicator Aalysis (TITAN) revealed the taxa and traits which exhibited the greatest changes in abundance or occurrence probability across gradients of fine sediment deposition and infiltration. Gradient forest analysis supported the TITAN results, showing that the greatest rate of turnover occurred at the lowest levels of fine sediment. The output of this analysis also highlighted shredders as the most responsive trait, supporting the conclusions of Chapter 3 which showed that shredders are consistently sensitive to fine sediment.

To conclude, addressing this aim was achieved through a variety of methodological approaches. Each chapter in this thesis demonstrates a distinct methodological approach which results in a distinct yet complimentary set of conclusions. It was essential that this diverse approach was employed in order to achieve this aim, and to gain a holistic view of this complex problem whilst providing a novel contribution to knowledge.

# 6.2.2 Aim 2 - Compare and assess methods for quantifying suspended and deposited fine sediment in lowland gravel bed rivers

Chapter 2 outlines traditional methods of measuring fine sediment which, while useful, can be time consuming, prone to errors and fail to integrate the

conditions of the catchment, often only representing conditions at a single point in time. Furthermore, there is no globally agreed standard practice, and the multitude of methods available each measure a different component of the fine sediment system. Considering the move towards the use of sediment-specific biomonitoring tools, the development of biomonitoring indices requires full testing of the pressure-response (Birk et al. 2012). The two sediment-specific indices which have been developed for use in the UK each used contrasting methods of measuring fine sediment in their development. EPSI (Turley et al. 2015, 2016) uses visual estimates, whereas CoFSI (Murphy et al. 2015) used the resuspension method. Critique of EPSI has focused around the lack of accuracy of the visual estimate method (Murphy et al. 2015). Visual estimates of fine sediment are susceptible to high user variability, and can either under- or over- estimate the total subsurface sediments depending on the vertical stratification of particles in the subsurface layer (Bunte and Abt 2001, Duerdoth et al. 2015). To achieve this aim, visual estimates, the resuspension method and SSC were assessed in Chapter 5.

Chapter 5 built on recommendations made by Conroy et al. (2016b) who recommended further testing of these fine sediment metrics in field conditions. The results presented show that visual estimates and total surface sediment (from the resuspension method) are strongly correlated. Visual estimates are a quick and instantaneous method of assessing fine sediment. The resuspension method requires more time investment and equipment making it unsuitable for routine monitoring. However, it is still useful for research purposes as it has the potential to yield additional information about the sediment quality than visual estimates alone. In addition to the metrics derived from the resuspension method within this thesis, sediment can also be tested for other parameters which may affect ecological responses such as particle size analysis and heavy metal content. This research has shown that visual estimates are a reliable proxy for fully quantitative measures when assessing fine sediment in lowland gravel bed rivers. As inter-operator variability was eliminated in the current study, methods for improving accuracy could be adopted in future studies. Clapcott et al. (2011) developed a method using a bathyscope to observe the

percentage of fine sediment within the gridded viewing area of the scope. The method is accompanied by a toolkit which provides images of examples of varying percentages of fine sediment in different substrate compositions. This aims to improve accuracy by allowing the operator to use these existing examples for comparison whilst carrying out the procedure. More time consuming methods include the digital image analysis developed by Turley et al. (2017), which is potentially highly accurate however requires significantly longer processing times.

# 6.2.3 Aim 3 - Test the response of macroinvertebrates to different metrics of fine sediment

When addressing this aim, it was vitally important that new independent data were collected in order to impartially test recently developed sediment-specific indices which both have the potential to be integrated in to national monitoring and assessment practices. This aim was particularly focussed on the performance of the sediment-specific indices PSI and its derived index EPSI (Extence et al. 2013, Turley et al. 2015, 2016), as well as CoFSI (Murphy et al. 2015), whilst also reviewing the performance of non-specific indices. Considering the current political climate in the UK (see Chapter 1, Section 1.1) and the opportunity to influence future monitoring and assessment approaches, it is important to understand the best way in which these tools can be used by both the Environment Agency and academics. Chapter 5 represents the first independent study to test the performance of both of these sediment-specific indices along with indices of general aquatic health in the UK.

In order to test the sediment-specific index performance in relation to fine sediment stress, a rigorous screening process was carried out during field site selection in order to minimise the impact of confounding variables. Once sites were selected, standard field and laboratory protocols (e.g. kick-net sampling, mixed taxa identification etc.) were followed so that the results would be compatible with current national monitoring practices. The results showed a strong correlation between PSI derived indices and fine sediment. EPSImixed was the best performing index which incorporates both a mechanistic basis

(sensitivity scores assigned based on expert knowledge) and an empirical element (weightings based on taxa abundance/sediment data) and has been most recently converted to mixed taxon level to be consistent with the standard practices of the Environment Agency. Overall, EPSImixed was the best performing index. The low number of scoring taxa, the removal of potentially sensitive species during index development, and the presence/absence nature of the scoring system, combined to limit the performance of CoFSI within this study. All non-specific indices tested performed less well, indicating the utility of sediment-specific indices. However, the close relationship between organic metrics and the sediment-specific indices demonstrate the congruence between organic and fine sediment pressures. Antecedent flow metrics were highly predictive of most indices, indicating the close link between flow, sediment transport and the other abiotic conditions which they influence. The ability of any sediment-specific index to efficiently identify sediment stress is impressive given the environmental variables which dominate community composition and the relatively low 'signal' of fine sediment pressures. Chapter 5 effectively tested the current sediment-specific indices with potential for use in the UK and is able to use the results to make recommendations for national monitoring bodies.

### 6.3 Implications for research

This thesis has built upon the work of previous publications which have sought to identify the most suitable macroinvertebrate indices for indicating fine sediment pressure. In a similar study Conroy et al. (2016a) collected field data of macroinvertebrates and multiple fine sediment metrics. Generalised linear mixed-effects models were used to determine which macroinvertebrate index was best explained by sediment metrics (surface cover from visual estimates, total surface sediment from resuspension samples, and turbidity). The results of Conroy et al. (2016a) showed that %EPT abundance (percentage of families from Ephemeroptera, Plecoptera and Trichoptera) had the strongest relationship with fine sediment, whereas all trait-based indices were poorly related. By applying different modelling methods, and including other abiotic

variables in the analysis, this thesis has teased apart the additional factors which contribute to the variance of both sediment metrics and biotic indices (and the relationships between them). The results of the models in this study have a much higher goodness-of-fit (R<sup>2</sup>) than that of Conroy et al. (2016a), highlighting the importance of considering the wider impact of the environment on index performance.

This thesis tested two distinct sediment-specific biomonitoring approaches which have been developed for use in the UK. However, there are multiple other approaches currently being developed globally. Gieswein, Hering, and Lorenz (2019), recently developed a fine sediment-specific biotic index using species sensitivity scores derived from TITAN analysis. Notably, Gieswein, Hering, and Lorenz (2019) suggest that biomonitoring indices need to be site specific. This is contradictory to the ambition to create nationally applicable indices (Environment Agency, *pers comm*).

The results of this thesis support the potential for trait-based monitoring by confirming some consistent trait responses. Trait-based approaches do not rely on taxonomic identity and are therefore not constrained by biogeographical variations across regions. For example, Doretto et al. (2018) compared the performance of 12 candidate indices (a mixture of both taxa- and trait- based) to gradients of fine sediment. The results showed that a combination of the best performing indices was the most effective biomonitoring approach. The resulting Multi-Metric Index (MMI) was based on indices which did not rely on species identity and therefore has the promise to be more spatially relevant. However, the MMI calibration was carried out using taxa collected from sediment traps at only one site. Organisms found in the sediment traps after removal may be dominated by stronger colonisers, rather than reflecting their sensitivities to fine sediment.

It is undoubtedly important to consider processes acting at different spatial scales when understanding the effects of fine sediment on macroinvertebrates and the potential importance of metacommunities (Leibold et al. 2004). The work by both Conroy et al. (2016a) and Doretto et al. (2018), described above,

was conducted at the patch scale (microhabitat), whereas this thesis has focused on macroinvertebrate responses at the reach (i.e. standard macroinvertebrate kick sampling area; Figure 5.4) scale. A closer relationship between taxa/trait responses to abiotic conditions would be expected at the microhabitat scale. In most cases, biotic indices are developed at the reach scale which can hinder understanding of the relationship between fine sediment and macroinvertebrate response (Larsen, Vaughan, and Ormerod 2009). Lamouroux, Doledec, and Gayraud (2004; p449) noted that '*the functional variability of invertebrate communities in stream reaches depended largely on microhabitat filters but also on other filters prevailing at the reach and larger scale*'. It is therefore important that responses are understood at a variety of scales. This thesis has added valuable information at the reach scale.

This thesis has helped to pick apart the interactions between taxa-traitenvironment relationships and responses to fine sediment. In Chapter 2, the emergence of trait-based approaches in ecology were discussed. The use of traits can have several advantages over traditional approaches. For example, trait-based approaches can transcend boundaries in taxonomic distributions between regions (Lancaster, Downes, and Glaister 2009), can avoid over or under emphasis of abundant species (Townsend and Hildrew 1994, Verberk, van Noordwijk, and Hildrew 2013), and provide a greater mechanistic understanding of the interactions between the environment and the ecological community (i.e. taxa-trait-environment interactions) (Pilière et al. 2016, Doretto et al. 2018). However, selection pressures do not act on individual traits, but on species whose combination of traits (or 'trait syndrome') controls their success in a particular environment (Verberk, van Noordwijk, and Hildrew 2013, Pilière et al. 2016). Due to this complexity, consistent individual trait-environment relationships are rare (Statzner and Bêche 2010), although through two distinct analyses (Chapter 3 and Chapter 5) this thesis has confirmed the consistency of shredders as sensitive to fine sediment. The FD indices used in this thesis, FDis and FRic, were not significantly related to any metrics of fine sediment. Additionally, they were poorly predicted by abiotic variables in linear models. It is clear that trait-based approaches need further development through either

development of the trait-based indices or the trait databases themselves. Wilkes et al. (2017) recommended a refined set of traits specifically for fine sediment biomonitoring. Examples of refinements or new trait modalities include: the ability or potential to excavate in the event of fine sediment burial (see Table 2.2); splitting the filter-feeding trait modality into those that can excrete excess fine sediment; and, behavioural or anatomical adaptations allowing gill respiration in highly sedimented environments. The results of Chapter 4 indicated that varying gill type could affect macroinvertebrate sensitivity to fine sediment, therefore gill type could be included as a new trait category. Better understanding of the mechanistic responses of macroinvertebrates to fine can help improve the trait databases.

## 6.4 Implications for sediment monitoring and assessment

The results of this thesis point to the use of biomonitoring indices as useful tools for sediment-specific monitoring in river management. Specifically, EPSImixed (Turley et al. 2016) was the best performing ecological indicator of fine sediment. Therefore, EPSImixed is the most appropriate biomonitoring index to be adopted into national monitoring and assessment practices. CoFSI needs further refinement before it can be applied in the same way.

Typically, when the intensity of a single-pressure acting on an ecological community is moderate to low, macroinvertebrates will exhibit a 'wedge-like' response, as discussed by Friberg (2010) (Figure 6.1). At low and moderate pressure, the interactions of the environment (abiotic and biotic) will dominate. This creates problems the use of single-metrics to detect single-pressures. The results of this thesis support this theory. In Chapter 5, the gradient forest analysis showed highest turnover in low to moderate sediment pressure. Friberg (2010) recommends a multi-metric approach in order to increase accuracy across the entire pressure gradient in order to counteract these interactions which act as a hindrance to single-metric approaches (e.g. Archaimbault et al. 2009). This thesis supports the idea that biomonitoring indices should not be used indiscriminately (i.e. in isolation). Using a suite of

indices (i.e. Figures 5.16 and 5.17) and observing the deviations between pressure/response relationships can help improve our understanding (Extence et al. 2017). In a multi-stressor environment, the use of pressure-specific indices can help to identify the hierarchy of stressors, and therefore where mitigation measures should be prioritised.

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Figure 6.1 – Typical 'wedge-shape' response indicative of single pressureresponse relationships (Friberg 2010).

The development of environmental DNA approaches (eDNA) provides the potential to revolutionise standard ecological assessments (Baird and Hajibabaei 2012). Water, soil or other substrates can be filtered to extract DNA which is then sequenced. The DNA sequences are then matched against known reference libraries. This method is non-invasive, has high detection capabilities and can provide rapid estimates of species presence compared to kick net-sampling and accompanying species level identification by taxonomists (Pawlowski et al. 2018). Current eDNA techniques can only indicate a species presence which will ultimately limit its predictive power. Attempts have been made to estimate species abundance by linking the number of DNA sequences to biomass, yet this is influenced by many factors including biomass, flow, acidity, UV radiation, species identity and life-stage (Spear et al. 2015, Goldberg et al. 2016). This thesis has shown the relatively poorer performance

of presence/absence only indices (e.g. CoFSI) compared to those which incorporate abundance (e.g. PSI and EPSI). Therefore, it is unlikely that eDNA can provide a direct replacement for current ecological assessment practices at present, although it does have the potential to act as a complimentary tool (Rees et al. 2014b). For example, in the use of detection of invasive species (e.g. the invasive crayfish *Oronectes rusticus*; Dougherty et al. 2016) or rare and threatened species (e.g. the Great Crested Newt; Rees et al. 2014a). Future applications could also include aspects of aquatic systems not currently monitored, such as bacteria and fungi, allowing a more holistic approach to assessment.

The invasion of non-native species is one of the main drivers of biodiversity change globally (Sala et al. 2000, Simberloff et al. 2013). Invasive non-native species can have far-reaching ecological effects and pose a challenge to monitoring practices (e.g. Dikerogammarus villosus; Macneil et al. 2013). For example, the invasive signal crayfish (*Pacifastacus leniusculus*) burrows in to river banks, resuspending sediment, destabilising riverbanks and increasing erosion (Rice et al. 2014, Holdich et al. 2014). In addition to its zoogeomorphic activity, selective predation may significantly affect the macroinvertebrate community (Parkyn, Rabeni, and Collier 1997, Usio et al. 2006). There is some evidence suggesting that *P. leniusculus* can artificially inflate PSI scores (Mathers et al. 2016). This could potentially lead to a false negative, where sites affected by fine sediment are artificially high PSI scores (i.e. low fine sediment condition) as a result of predation on sediment-tolerant species. Although invasive species have not been considered in this thesis the most recent evidence by Turley et al. (2017b) showed that whilst EPSI (the empirically developed PSI index) did show small changes pre- and post-invasion by P. *leniusculus*, its presence is unlikely to result in incorrect diagnoses of sediment pressure. Considering each year 10-12 new non-native species become established in the UK, it is important to continually consider the effect these species may have on biomonitoring indices (Defra 2015).
## 6.5 Future priorities

This thesis has identified the urgent need to prioritise the development of the mechanistic understanding of macroinvertebrate responses to fine sediment through the production of high-quality research. It is important that future research utilises a variety of theoretical approaches such as reductionist mechanistic style studies (rather than the current trend of correlative field-based research). Fine sediment particle size must be reported as a minimum requirement when disseminating research through academic publications. The results of the systematic review (Chapter 3) showed that this is not standard practice. Furthermore, an international definition of fine sediment should be adopted. It is recommended that this definition sits in line with the grain size categories outlined in the River Habitat Survey (Table 2.4). These also correspond to the size categories outlined in the Wentworth scale (Wentworth 1922) which has been in use for nearly 100 years. Further development is required focussing on new and emerging methods of studying multiple trait responses rather than focusing on individual trait responses, which have already been studied extensively and show few consistent relationships. Incorporating functional trait niches, trait groups or life history strategies can help improve the understanding of trait-environment relationships (Poff et al. 2006, Verberk, van Noordwijk, and Hildrew 2013).

Developing on the potential for further trait research, when assigning taxa with a sensitivity score or weighting in biotic index development, an organism's functional niche must be considered. The niche that a species occupies at a specific site is dependent on the prevailing abiotic and biotic conditions. Therefore, the niche a particular species occupies, and the resources that it uses, is affected by the niches occupied by other taxa in an environment (and the resources that they use). This is known as niche complementarity (Ashton et al. 2010). Any adjustment to this, such as impact by a stressor like fine sediment, will have cascading effects across all trait groups in the environment. These interactions are difficult to capture in biomonitoring indices and can be largely site specific. This is why patterns from single site studies can be difficult to extrapolate more generally. Linked with niche complementarity is the theory

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of functional redundancy. Functional redundancy is the concept of multiple species providing similar roles within the same community. Ecosystems with higher functional redundancy will be more resilient (Schmera et al. 2017), but functional redundancy is difficult to accurately measure (Solow and Polasky 1994, Bady et al. 2005). By better understanding these intricacies through further research, we can more reliably capture trait responses, improving monitoring practices and also contributing to the global understanding of ecosystem functioning.

The future of biomonitoring indices cannot be discussed without the consideration of their performance in a changing climate. The current RICT model uses abiotic variables which are less predictable in a changing climate (e.g. temperature and flow category). Using the current model to predict expected index scores (which are used in reporting on the current WFD as Ecological Quality Ratios) will need to be updated to reflect variations in temperature and precipitation under climate change. The UK Climate Change Projection 2018 (UKCP18) (Lowe et al. 2018) predicts winter rainfall to increase and summer rainfall to decrease but storm events to become more frequent and intense. This will result in significant effects on river flow (hydrological metrics) as well as the erosion, transport and deposition of fine sediments in river systems. Notwithstanding the potential effects of warming temperatures on macroinvertebrates, monitoring networks require regular fitness checks to ensure the effects of a changing climate can be adequately reflected (Wilby et al. 2010, Reid et al. 2019). Beyond the potential direct impacts of climate change in aquatic systems, Floury et al. (2017) notes that improved water quality management can significantly help to reduce some adverse effects of climate change, further emphasising the need for robust monitoring practices.

This thesis has shown the close link between flow and sediment-specific indices as well as indices for general aquatic health. When considering the future development of biomonitoring practices, it is important to consider the key interactions between flow conditions and community assembly processes. Both river longitudinal slope, discharge, and therefore stream power, change as distance downstream increases. This will have a direct influence on

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macroinvertebrate community structure (when considering hydraulic habitat preferences) but indirect effects via fine sediment transport and storage. Additionally, at mid-basin locations, species composition exhibits high rates of turnover and rapid sorting and selection of adapted taxa from the species pool (Wilkes et al. 2019). This is compared to headwaters where colonisation is limited and further downstream where the effects of stressors are drowned out by an influx of organisms from the river network upstream (Poff 1997, Leibold et al. 2004). Mid-basin locations (or mid-order streams) therefore represent a pivotal condition of both stream power and demographic biotic processes. These conditions potentially contribute to the optimum performance of any biotic index (i.e. index optimisation; Figure 6.2). There is potential for this theory to be developed in the context of the application of sediment-specific indices.

## 6.6 Concluding remarks

Fine sediment is a significant pressure on freshwater systems globally. Management interventions to identify sources of fine sediment are dependent on appropriate monitoring methods. Aquatic macroinvertebrates are widely used as biomonitors in river systems. This thesis has investigated the response of macroinvertebrates to fine sediment in order to inform biomonitoring practices. This thesis contains the first systematic-style review of macroinvertebrate responses to fine sediment which map the breadth of existing evidence and highlights significant knowledge gaps. The results have emphasised the need for a better understanding of the mechanisms behind macroinvertebrate responses to fine sediment. Through identifying a particular knowledge gap, this thesis determined the potential for physical damage by fine sediment on macroinvertebrate gills. The results of this thesis support the potential for sediment-specific indices, in particular EPSImixed was the best performing index and its ability to detect fine sediment stress is remarkable given the potentially confounding effects of myriad other environmental variations.





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*Ecohydraulics*. Chichester, UK, 7–30

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Zweig, L.D. and Rabeni, C.F. (2001) 'Biomonitoring for Deposited Sediment Using Benthic Invertebrates: A Test on 4 Missouri Streams'. *Journal of the North American Benthological Society* 20 (4), 643–657

## **Appendix 1**

### Appendix 1.1 Scopus search link for evidence review

https://www.scopus.com/results/results.uri?numberOfFields=0&src=s&clickedLi nk=&edit=&editSaveSearch=&origin=searchbasic&authorTab=&affiliationTab=& advancedTab=&scint=1&menu=search&tablin=&searchterm1=invertebrates+O R+macroinvertebrates+AND+sediment+OR+fine+sediment+OR+sand+OR+silt+ OR+clay+OR+colloid&field1=TITLE\_ABS\_KEY&dateType=Publication\_Date\_T ype&yearFrom=Before+1960&yearTo=Present&loadDate=7&documenttype=All &resetFormLink=&st1=invertebrates+OR+macroinvertebrates+AND+sediment+ OR+fine+sediment+OR+sand+OR+silt+OR+clay+OR+colloid&st2=&sot=b&sdt= b&sl=115&s=TITLE-ABS-KEY%28invertebrates+OR+macroinvertebrates+AND+sediment+OR+fine+sedi ment+OR+sand+OR+silt+OR+colloid%29&sid=F1082AA4013D7D5C 0837BCB8C7E30609.wsnAw8kcdt7IPYLO0V48qA%3A10&searchId=F1082AA

4013D7D5C0837BCB8C7E30609.wsnAw8kcdt7IPYLO0V48gA%3A10&txGid=F 1082AA4013D7D5C0837BCB8C7E30609.wsnAw8kcdt7IPYLO0V48gA%3A1&s ort=plf-f&originationType=b&rr=

### Appendix 1.2 Articles included in the evidence review

Table A1.1 – Bibliographic information for all articles included in the evidence review. \*Where sediment says was not quantified in the original article, the sediment size is described as 'not recorded'.

Full reference	Sediment	Sediment size	Country	Study type
	fraction			
Albertson, L.K., Daniels, M.D. (2016). Resilience of aquatic net-spinning	Deposited	<355 µm	USA	Experimental
caddisfly silk structures to common global stressors. Freshwater	and			
Biology 61(5), 670-679	suspended			
Angradi, T.R. (1999). Fine sediment and macroinvertebrate	Deposited	<2 mm	USA	Both
assemblages in Appalachian streams: A field experiment with				
biomonitoring applications. Journal of the North American Benthological				
Society 18(1), 49-66				
Aspray K.L., Holden J., Ledger M.E., Mainstone C.P., Brown L.E.	Deposited	Not recorded	UK	Experimental
(2017). Organic sediment pulses impact rivers across multiple levels of				
ecological organization. Ecohydrology 10(6), e1855				
Beermann A.J., Elbrecht V., Karnatz S., Ma L., Matthaei C.D., Piggott	Deposited	<500 µm	Germany	Experimental
J.J., Leese F. (2018). Multiple-stressor effects on stream				
macroinvertebrate communities: A mesocosm experiment manipulating				
salinity, fine sediment and flow velocity. Science of the Total				
Environment 610-611(), 961-971				
Béjar M., Gibbins C.N., Vericat D., Batalla R.J. (2017). Effects of	Suspended	Not recorded	Spain	Observational
Suspended Sediment Transport on Invertebrate Drift. River Research				
and Applications 33(10), 1655-1666				
Benoy, G.A., Sutherland, A.B., Culp, J.M., Brua, R.B. (2012). Physical	Deposited	<2 mm	Canada	Observational
and ecological thresholds for deposited sediments in streams in				
agricultural landscapes. Journal of Environmental Quality 41(1), 31-40				

Full reference	Sediment	Sediment size	Country	Study type
	fraction			
Berkman, H.E., Rabeni, C.F., Boyle, T.P. (1986). Biomonitors of stream	Deposited	<0.5 mm	USA	Observational
quality in agricultural areas: Fish versus invertebrates. Environmental				
Management 10(3), 413-419				
Blettler, M.C.M., Amsler, M.L., Ezcurra De Drago, I., Drago, E., Paira,	Suspended	Not recorded	Paraguay-	Observational
A., Espinola, L.A., Eberle, E., Szupiany, R. (2016). Fine sediment input		(SSC only)	Argentina	
and benthic fauna interactions at the confluence of two large rivers.				
International Journal of Environmental Research 10(1), 65-76				
Blettler, M.C.M., Amsler, M.L., Ezcurra de Drago, I., Espinola, L.A.,	Suspended	Not recorded	Paraguay-	Observational
Eberle, E., Paira, A., Best, J.L., Parsons, D.R., Drago, E.E. (2015). The		(SSC only)	Argentina	
impact of significant input of fine sediment on benthic fauna at tributary				
junctions: A case study of the Bermejo-Paraguay River confluence,				
Argentina. Ecohydrology 8(2), 340-352				
Bo, T., Fenoglio, S., Malacarne, G., Pessino, M., Sgariboldi, F. (2007).	Deposited	0.25-0.5 mm	Italy	Experimental
Effects of clogging on stream macroinvertebrates: An experimental				
approach. Limnologica 37(2), 186-192				
Bona, F., Doretto, A., Falasco, E., La Morgia, V., Piano, E., Ajassa, R.,	Deposited	<0.105 mm	Italy	Observational
Fenoglio, S. (2016). Increased Sediment Loads in Alpine Streams: An	and			
Integrated Field Study. River Research and Applications 32(6), 1316-	suspended			
1326				
Bond, N.R., Downes, B.J. (2003). The independent and interactive	Suspended	500-1000 µm	Australia	Experimental
effects of fine sediment and flow on benthic invertebrate communities				
characteristic of small upland streams. Freshwater Biology 48(3), 455-				
465				

Full reference	Sediment	Sediment size	Country	Study type
	fraction			
Braccia, A., Voshell Jr., J.R. (2006). Environmental factors accounting	Deposited	<2 mm	USA	Observational
for benthic macroinvertebrate assemblage structure at the sample scale				
in streams subjected to a gradient of cattle grazing. Hydrobiologia				
573(1), 55-73				
Broekhuizen, N., Parkyn, S., Miller, D. (2001). Fine sediment effects on	Deposited	<63 µm	New	Experimental
feeding and growth in the invertebrate grazers Potamopyrgus			Zealand	
antipodarum (Gastropoda, Hydrobiidae) and Deleatidium sp.				
(Ephemeroptera, Leptophlebiidae). Hydrobiologia 457(), 125-132				
Buendia, C., Gibbins, C.N., Vericat, D., Batalla, R.J. (2014). Effects of	Deposited	<2 mm	Spain	Observational
flow and fine sediment dynamics on the turnover of stream invertebrate				
assemblages. Ecohydrology 7(4), 1105-1123				
Buendia, C., Gibbins, C.N., Vericat, D., Batalla, R.J., Douglas, A.	Deposited	<2 mm	Spain	Observational
(2013). Detecting the structural and functional impacts of fine sediment				
on stream invertebrates. Ecological Indicators 25(), 184-196				
Burdon, F.J., McIntosh, A.R., Harding, J.S. (2013). Habitat loss drives	Deposited	<2 mm	New	Observational
threshold response of benthic invertebrate communities to deposited			Zealand	
sediment in agricultural streams. Ecological Applications 23(5), 1036-				
1047				
Chase J.W., Benoy G.A., Culp J.M. (2017). Combined effects of nutrient	Deposited	<2 mm	Canada	Experimental
enrichment and inorganic sedimentation on benthic biota in an				
experimental stream system. Water Quality Research Journal of				
Canada 52(3), 151-165				
Chiu, MC., Yeh, CH., Sun, YH., Kuo, MH. (2013). Short-term	Deposited	Not recorded	Taiwan	Observational
effects of dam removal on macroinvertebrates in a Taiwan stream.				
Aquatic Ecology 47(2), 245-252				

Full reference	Sediment	Sediment size	Country	Study type
	fraction			
Ciesielka, I.K., Bailey, R.C. (2001). Scale-specific effects of sediment	Deposited	<200 µm	Canada	Experimental
burial on benthic macroinvertebrate communities. Journal of Freshwater				
Ecology 16(1), 73-81				
Connolly, N.M., Pearson, R.G. (2007). The effect of fine sedimentation	Deposited	<200 µm	Australia	Experimental
on tropical stream macroinvertebrate assemblages: A comparison using	and			
flow-through artificial stream channels and recirculating mesocosms.	suspended			
Hydrobiologia 592(1), 423-438				
Conroy E., Turner J.N., Rymszewicz A., Bruen M., O'Sullivan J.J.,	Deposited	four size	Ireland	Experimental
Lawler D.M., Stafford S., Kelly-Quinn M. (2018). Further insights into		classes - all <2		
the responses of macroinvertebrate species to burial by sediment.		mm		
Hydrobiologia 805(1), 399-411				
Conroy, E., Turner, J.N., Rymszewicz, A., Bruen, M., O'Sullivan, J.J.,	Deposited	<1 mm	Ireland	Both
Lawler, D.M., Lally, H., Kelly-Quinn, M. (2016). Evaluating the	and			
relationship between biotic and sediment metrics using mesocosms and	suspended			
field studies. Science of the Total Environment 568(), 1092-1101				
Couceiro, S.R.M., Hamada, N., Forsberg, B.R., Padovesi-Fonseca, C.	Suspended	Not recorded	Brazil	Observational
(2011). Trophic structure of macroinvertebrates in Amazonian streams		for SSC and		
impacted by anthropogenic siltation. Austral Ecology 36(6), 89-103		<63 µm for the		
		deposited		
		sediment		
Couceiro, S.R.M., Hamada, N., Forsberg, B.R., Padovesi-Fonseca, C.	Suspended	Not recorded	Brazil	Observational
(2009). Effects of anthropogenic silt on aquatic macroinvertebrates and		for SSC and		
abiotic variables in streams in the Brazilian Amazon. Journal of Soils		<63 µm for the		
and Sediments 10(1), 89-103		deposited		
		sediment		

Full reference	Sediment	Sediment size	Country	Study type
	fraction			
Cover, M.R., May, C.L., Dietrich, W.E., Resh, V.H. (2008). Quantitative	Deposited	4 mm	USA	Observational
linkages among sediment supply, streambed fine sediment, and benthic				
macroinvertebrates in northern California streams. Journal of the North				
American Benthological Society 27(1), 135-149				
Culp, J.M., Brua, R.B., Benoy, G.A., Chambers, P.A. (2012).	Suspended	Not recorded	Canada	Observational
Development of reference conditions for suspended solids in streams.		for SSC		
Canadian Water Resources Journal 38(2), 85-98				
Culp, J.M., Wrona, F.J., Davies, R.W. (1986). Response of stream	Deposited	0.5-2 mm sand	Canada	Experimental
benthos and drift to fine sediment deposition versus transport.	and	particles		
Canadian Journal of Zoology 64(6), 1345-1351	suspended			
Dabney B.L., Clements W.H., Williamson J.L., Ranville J.F. (2018).	Deposited	<2360 µm	USA	Experimental
Influence of Metal Contamination and Sediment Deposition on Benthic				
Invertebrate Colonization at the North Fork Clear Creek Superfund Site,				
Colorado, USA. Environmental Science and Technology 52(12), 7072-				
7080				
Davis S.J., Ó hUallacháin D., Mellander PE., Kelly AM., Matthaei	Deposited	< 500 µm	Ireland	Experimental
C.D., Piggott J.J., Kelly-Quinn M. (2018). Multiple-stressor effects of				
sediment, phosphorus and nitrogen on stream macroinvertebrate				
communities. Science of the Total Environment 637-638(), 577-587				
Davis, A.M., Pearson, R.G., Kneipp, I.J., Benson, L.J., Fernandes, L.	Deposited	Not recorded	Australia	Observational
(2015). Spatiotemporal variability and environmental determinants of				
invertebrate assemblage structure in an Australian dry-tropical river.				
Freshwater Science 34(2), 634-647				

Full reference	Sediment fraction	Sediment size	Country	Study type
De Castro Vasconcelos, M., Melo, A.S. (2008). An experimental test of the effects of inorganic sediment addition on benthic macroinvertebrates of a subtropical stream. Hydrobiologia 610(1), 321- 329	Suspended	fine sand 0- 0.24 mm, coarse sand 0.25-0.8 mm	Brazil	Experimental
De Drago, I.E., Marchese, M., Wantzen, K.M. (2004). Benthos of a large neotropical river: Spatial patterns and species assemblages in the Lower Paraguay and its floodplains. Archiv fur Hydrobiologie 160(3), 347-374	Deposited	various - doesn't classify	Paraguay- Argentina	Observational
Descloux, S., Datry, T., Marmonier, P. (2013). Benthic and hyporheic invertebrate assemblages along a gradient of increasing streambed colmation by fine sediment. Aquatic Sciences 75(4), 493-507	Deposited	<2 mm	France	Both
Descloux, S., Datry, T., Usseglio-Polatera, P. (2014). Trait-based structure of invertebrates along a gradient of sediment colmation: Benthos versus hyporheos responses. Science of the Total Environment 466-467(), 265-276	Deposited	particles <2 mm	France	Observational
Doeg, T.J., Milledge, G.A. (1991). Effect of experimentally increasing concentrations of suspended sediment on macroinvertebrate drift. Marine and Freshwater Research 42(5), 519-526	Suspended	Clay	Australia	Experimental
Dolédec, S., Phillips, N., Scarsbrook, M., Riley, R.H., Townsend, C.R. (2006). Comparison of structural and functional approaches to determining landuse effects on grassland stream invertebrate communities. Journal of the North American Benthological Society 25(1), 44-60	Deposited	particles <1 mm	New Zealand	Observational
Doretto A., Bona F., Piano E., Zanin I., Eandi A.C., Fenoglio S. (2017). Trophic availability buffers the detrimental effects of clogging in an alpine stream. Science of the Total Environment 592(), 503-511	Deposited	<1 mm	Italy	Experimental

Full reference	Sediment	Sediment size	Country	Study type
	fraction			
Doretto, A., Bona, F., Falasco, E., Piano, E., Tizzani, P., Fenoglio, S.	Deposited	<0.105 mm	Italy	Experimental
(2016). Fine sedimentation affects CPOM availability and shredder				
abundance in Alpine streams. Journal of Freshwater Ecology 31(2),				
299-302				
Duan, X., Wang, Z., Tian, S. (2008). Effect of streambed substrate on	Deposited	fine sand (0.2	China	Experimental
macroinvertebrate biodiversity. Frontiers of Environmental Science and		mm) and		
Engineering in China 2(1), 122-128		coarse sand		
		(1.5 mm)		
Elbrecht, V., Beermann, A.J., Goessler, G., Neumann, J., Tollrian, R.,	Deposited	<0.5 mm (500	Germany	Experimental
Wagner, R., Wlecklik, A., Piggott, J.J., Matthaei, C.D., Leese, F. (2016).		µm)		
Multiple-stressor effects on stream invertebrates: A mesocosm				
experiment manipulating nutrients, fine sediment and flow velocity.				
Freshwater Biology 61(4), 362-375				
Espa, P., Castelli, E., Crosa, G., Gentili, G. (2013). Environmental	Deposited	Not recorded	Italy	Observational
effects of storage preservation practices: Controlled flushing of fine	and			
sediment from a small hydropower reservoir. Environmental	suspended			
Management 52(1), 261-276				
Everall N.C., Johnson M.F., Wood P., Mattingley L. (2018). Sensitivity	Suspended	Not recorded	UK	Experimental
of the early life stages of a mayfly to fine sediment and orthophosphate				
levels. Environmental Pollution 237(), 792-802				
Forio M.A.E., Lock K., Radam E.D., Bande M., Asio V., Goethals P.L.M.	Suspended	Not recorded	Phillipines	Observational
(2017). Assessment and analysis of ecological quality,				
macroinvertebrate communities and diversity in rivers of a				
multifunctional tropical island. Ecological Indicators 77(), 228-238				

Full reference	Sediment	Sediment size	Country	Study type
	fraction			
Fossati, O., Wasson, JG., Héry, C., Salinas, G., Marín, R. (2001).	Suspended	Not recorded	Bolivia	Observational
Impact of sediment releases on water chemistry and macroinvertebrate				
communities in clear water Andean streams (Bolivia). Archiv fur				
Hydrobiologie 151(1), 33-50				
Fritz, K.M., Dodds, W.K., Pontius, J. (1999). The effects of bison	Deposited	<0.2 mm	USA	Observational
crossings on the macroinvertebrate community in a tallgrass prairie	and	suspended and		
stream. American Midland Naturalist 141(2), 253-265	suspended	<2 mm		
		deposited		
García Molinos, J., Donohue, I. (2011). Temporal variability within	Deposited	<2 mm	Ireland	Experimental
disturbance events regulates their effects on natural communities.	and			
Oecologia 166(3), 1794-1800	suspended			
García Molinos, J., Donohue, I. (2010). Interactions among temporal	Deposited	<2 mm	Ireland	Experimental
patterns determine the effects of multiple stressors. Ecological				
Applications 20(7), 1794-1800				
Gayraud, S., Philippe, M. (2003). Influence of bed-sediment features on	Deposited	silt and clay (0	France	Observational
the interstitial habitat available for macroinvertebrates in 15 French		- 0.05 mm),		
streams. International Review of Hydrobiology 88(1), 667-686		sand (0.05 – 2		
		mm)		
Gayraud, S., Philippe, M. (2001). Does subsurface interstitial space	Deposited	<2 mm	France	Observational
influence general features and morphological traits of the benthic				
macroinvertebrate community in streams?. Archiv fur Hydrobiologie				
151(4), 667-686				
Glendell, M., Extence, C., Chadd, R., Brazier, R.E. (2014). Testing the	Deposited	<0.06 mm	UK	Observational
pressure-specific invertebrate index (PSI) as a tool for determining				
ecologically relevant targets for reducing sedimentation in streams.				
Freshwater Biology 59(2), 353-367				

Full reference	Sediment	Sediment size	Country	Study type
	fraction			
Gomi, T., Kobayashi, S., Negishi, J.N., Imaizumi, F. (2010). Short-term	Deposited	deposited >1	Japan	Experimental
responses of macroinvertebrate drift following experimental sediment	and	mm and		
flushing in a Japanese headwater channel. Landscape and Ecological	suspended	suspended <1		
Engineering 6(2), 257-270		mm		
Gordon, A.K., Griffin, N.J., Palmer, C.G. (2015). The relationship	Suspended	Not recorded	South	Observational
between concurrently measured sass (South African scoring system)			Africa	
and turbidity data archived in the South African river health				
programme's rivers database. Water SA 41(1), 21-26				
Graça, M.A.S., Ferreira, W.R., Firmiano, K., França, J., Callisto, M.	Deposited	<2 mm	Brazil	Both
(2015). Macroinvertebrate identity, not diversity, differed across patches				
differing in substrate particle size and leaf litter packs in low order,				
tropical Atlantic forest streams. Limnetica 34(1), 29-40				
Graeber D., Jensen T.M., Rasmussen J.J., Riis T., Wiberg-Larsen P.,	Deposited	Not recorded	Denmark	Experimental
Baattrup-Pedersen A. (2017). Multiple stress response of lowland				
stream benthic macroinvertebrates depends on habitat type. Science of				
the Total Environment 599-600(), 1517-1523				
Graf, W., Leitner, P., Hanetseder, I., Ittner, L.D., Dossi, F., Hauer, C.	Deposited	psammel	Austria	Observational
(2016). Ecological degradation of a meandering river by local		0.063–2 mm,		
channelization effects: a case study in an Austrian lowland river.		psammopelal		
Hydrobiologia 772(1), 145-160		<0.063 mm		
Gray, L.J., Ward, J.V. (1982). Effects of sediment releases from a	Suspended	Not recorded	USA	Observational
reservoir on stream macroinvertebrates. Hydrobiologia 96(2), 177-184				

Full reference	Sediment	Sediment size	Country	Study type
	fraction	0/		
Griffith, M.B., Daniel, F.B., Morrison, M.A., Troyer, M.E., Lazorchak,	Deposited	% sand = >	USA	Observational
J.M., Schubauer-Berigan, J.P. (2009). Linking excess nutrients, light,		0.06 – 2 mm,		
and fine bedded sediments to impacts on faunal assemblages in		% fines = <		
headwater agricultural streams. Journal of the American Water		0.06 mm -		
Resources Association 45(6), 1475-1492		these are		
		compined		
Growns I., Murphy J.F., Jones J.I. (2017). The effects of altered flow	Deposited	<2 mm	UK	Experimental
and bed sediment on macroinvertebrates in stream mesocosms. Marine				
and Freshwater Research 68(3), 496-505				
Hall Jr., L.W., Killen, W.D. (2005). Temporal and spatial assessment of	Deposited	Not recorded	USA	Observational
water quality, physical habitat, and benthic communities in an impaired	and			
agricultural stream in California's San Joaquin Valley. Journal of	suspended			
Environmental Science and Health - Part A Toxic/Hazardous				
Substances and Environmental Engineering 40(5), 959-989				
Harding, J.S., Jellyman, P.G. (2015). Earthquakes, catastrophic	Deposited	<500 µm	New	Observational
sediment additions and the response of urban stream communities.			Zealand	
New Zealand Journal of Marine and Freshwater Research 49(3), 346-				
355				
Harris, I.W.E., Drury, C.F., Simard, R.R., Zhang, T.Q. (2003). Density	Deposited	Sand = 0.2 – 2	Canada	Observational
and richness of benthic invertebrate populations in the North Sydenham		mm		
River of Southwestern Ontario (1996-2000) compared with those of the		Mud = < 0.2		
St. Clair River (1990-1995). Canadian Field-Naturalist 117(2), 267-277		mm		
Harrison, E.T., Norris, R.H., Wilkinson, S.N. (2008). Can an indicator of	Deposited	<5 mm	Australia	Observational
river health be related to assessments from a catchment-scale				
sediment model?. Hydrobiologia 600(1), 49-64				

Full reference	Sediment	Sediment size	Country	Study type
	fraction			
Hogg, I.D., Norris, R.H. (1991). Effects of runoff from land clearing and	Deposited	Deposited	Australia	Observational
urban development on the distribution and abundance of	and	sediments		
macroinvertebrates in pool areas of a river. Marine and Freshwater	suspended	<250 µm		
Research 42(5), 507-518				
Hubert, W.A., LaVoie IV, W.J., DeBray, L.D. (1996). Densities and	Deposited	<0.42 mm	USA	Observational
substrate associations of macroinvertebrates in riffles of a small, high				
plains stream. Journal of Freshwater Ecology 11(1), 21-26				
Hutchens Jr., J.J., Schuldt, J.A., Richards, C., Johnson, L.B., Host,	Deposited	Not recorded	USA	Observational
G.E., Breneman, D.H. (2009). Multi-scale mechanistic indicators of				
Midwestern USA stream macroinvertebrates. Ecological Indicators 9(6),				
1138-1150				
Jun, YC., Kim, NY., Kwon, SJ., Han, SC., Hwang, IC., Park, J	Deposited	<2 mm	Korea	Observational
H., Won, DH., Byun, MS., Kong, HY., Lee, JE., Hwang, SJ.	and			
(2011). Effects of land use on benthic macroinvertebrate communities:	suspended			
Comparison of two mountain streams in Korea. Annales de Limnologie				
47(), S35-S49				
Kaller, M.D., Hartman, K.J. (2004). Evidence of a threshold level of fine	Deposited	Mixed	USA	Observational
sediment accumulation for altering benthic macroinvertebrate				
communities. Hydrobiologia 518(43525), 95-104				
Kefford, B.J., Zalizniak, L., Dunlop, J.E., Nugegoda, D., Choy, S.C.	Deposited	Clay <63 µm	Australia	Experimental
(2010). How are macroinvertebrates of slow flowing lotic systems	and			
directly affected by suspended and deposited sediments?.	suspended			
Environmental Pollution 158(2), 543-550				
Kennedy, T.B., Merenlender, A.M., Vinyard, G.L. (2000). A comparison	Deposited	Not recorded	USA	Observational
of riparian condition and aquatic invertebrate community indices in				
central Nevada. Western North American Naturalist 60(3), 255-272				

Full reference	Sediment	Sediment size	Country	Study type
	fraction			
Kent, T.R., Stelzer, R.S. (2008). Effects of deposited fine sediment on	Deposited	<63 µm	USA	Experimental
life history traits of Physa integra snails. Hydrobiologia 596(1), 329-340				
Kreutzweiser, D.P., Capell, S.S., Good, K.P. (2005). Effects of fine	Deposited	<250 µm	Canada	Observational
sediment inputs from a logging road on stream insect communities: A				
large-scale experimental approach in a Canadian headwater stream.				
Aquatic Ecology 39(1), 55-66				
Lange, K., Townsend, C.R., Matthaei, C.D. (2014). Can biological traits	Deposited	<2 mm	New	Observational
of stream invertebrates help disentangle the effects of multiple stressors			Zealand	
in an agricultural catchment?. Freshwater Biology 59(12), 2431-2446				
Larsen, S., Ormerod, S.J. (2010). Combined effects of habitat	Deposited	<2 mm	UK	Observational
modification on trait composition and species nestedness in river				
invertebrates. Biological Conservation 143(11), 51-60				
Larsen, S., Ormerod, S.J. (2010). Low-level effects of inert sediments	Deposited	<2 mm	UK	Experimental
on temperate stream invertebrates. Freshwater Biology 55(2), 51-60				
Larsen, S., Pace, G., Ormerod, S.J. (2011). Experimental effects of	Deposited	0.2 - 1 mm	UK	Experimental
sediment deposition on the structure and function of macroinvertebrate				
assemblages in temperate streams. River Research and Applications				
27(2), 257-267				
Larsen, S., Vaughan, I.P., Ormerod, S.J. (2009). Scale-dependent	Deposited	<2 mm	UK	Observational
effects of fine sediments on temperate headwater invertebrates.				
Freshwater Biology 54(1), 203-219				
Lenat, D.R., Penrose, D.L., Eagleson, K.W. (1981). Variable effects of	Deposited	Not recorded	USA	Observational
sediment addition on stream benthos. Hydrobiologia 79(2), 187-194	and			
	suspended			

Full reference	Sediment	Sediment size	Country	Study type
	fraction			
Longing, S.D., Voshell Jr., J.R., Dolloff, C.A., Roghair, C.N. (2010).	Deposited	<2 mm	USA	Observational
Relationships of sedimentation and benthic macroinvertebrate				
assemblages in headwater streams using systematic longitudinal				
sampling at the reach scale. Environmental Monitoring and Assessment				
161(43556), 517-530				
Louhi, Pauliina Richardson, John S. Muotka, Timo (2017). Sediment	Deposited	<0.5 mm	Canada	Experimental
addition reduces the importance of predation on ecosystem functions in				
experimental stream channels. Canadian Journal of Fisheries & Aquatic				
Sciences 74(1), 32-40				
Lummer, EM., Auerswald, K., Geist, J. (2016). Fine sediment as	Suspended	<125 µm	Germany	Experimental
environmental stressor affecting freshwater mussel behavior and				
ecosystem services. Science of the Total Environment 571(), 1340-				
1348				
Magbanua, F.S., Townsend, C.R., Hageman, K.J., Piggott, J.J.,	Deposited	mean grain	New	Experimental
Matthaei, C.D. (2016). Individual and combined effects of fine sediment		size was 0.2	Zealand	
and glyphosate herbicide on invertebrate drift and insect emergence: A		mm		
stream mesocosm experiment. Freshwater Science 35(1), 139-151				
Magierowski, Regina H. Read, Steve M. Carter, Steven J. B. Warfe,	Deposited	<2 mm	Australia	Experimental
Danielle M. Cook, Laurie S. (2015). Inferring Landscape-Scale Land-	and			
Use Impacts on Rivers Using Data from Mesocosm Experiments and	suspended			
Artificial Neural Networks. PLoS ONE 10(3), e0120901				
Mary, N., Marmonier, P. (2000). First survey of interstitial fauna in New	Deposited	<2 mm	New	Observational
Caledonian rivers: Influence of geological and geomorphological			Caledonia	
characteristics. Hydrobiologia 418(1), 199-208				

Full reference	Sediment	Sediment size	Country	Study type
	fraction			
Mathers K.L., Rice S.P., Wood P.J. (2017). Temporal effects of	Deposited	<2 mm	UK	Experimental
enhanced fine sediment loading on macroinvertebrate community				
structure and functional traits. Science of the Total Environment 599-				
600(), 513-522				
Matthaei, C.D., Piggott, J.J., Townsend, C.R. (2010). Multiple stressors	Deposited	0.2 mm	New	Experimental
in agricultural streams: Interactions among sediment addition, nutrient			Zealand	
enrichment and water abstraction. Journal of Applied Ecology 47(3),				
639-649				
Matthaei, C.D., Weller, F., Kelly, D.W., Townsend, C.R. (2006). Impacts	Deposited	existing 2 mm -	New	Experimental
of fine sediment addition to tussock, pasture, dairy and deer farming		mean grain	Zealand	
streams in New Zealand. Freshwater Biology 51(11), 2154-2172		size of added		
		sediment is 0.2		
		mm		
Miliša, M., Živković, V., Habdija, I. (2010). Destructive effect of quarry	Suspended	Not recorded	Croatia	Observational
effluent on life in a mountain stream. Biologia 65(3), 520-526				
Miliša, M., Živković, V., Kepčija, R.M., Habdija, I. (2010). Siltation	Suspended	Not recorded	Croatia	Observational
disturbance in a mountain stream: Aspect of functional composition of				
the benthic community. Periodicum Biologorum 112(2), 173-178				
Molinos, J.G., Donohue, I. (2009). Differential contribution of	Suspended	<63 µm	Ireland	Experimental
concentration and exposure time to sediment dose effects on stream				
biota. Journal of the North American Benthological Society 28(1), 110-				
121				

Full reference	Sediment	Sediment size	Country	Study type
	fraction			
Murphy, J.F., Jones, J.I., Pretty, J.L., Duerdoth, C.P., Hawczak, A.,	Deposited	<2 mm	UK	Observational
Arnold, A., Blackburn, J.H., Naden, P.S., Old, G., Sear, D.A., Hornby,				
D., Clarke, R.T., Collins, A.L. (2015). Development of a biotic index				
using stream macroinvertebrates to assess stress from deposited fine				
sediment. Freshwater Biology 60(10), 2019-2036				
Mustonen, KR., Mykrä, H., Louhi, P., Markkola, A., Tolkkinen, M.,	Deposited	<2 mm	Finland	Experimental
Huusko, A., Alioravainen, N., Lehtinen, S., Muotka, T. (2016).				
Sediments and flow have mainly independent effects on multitrophic				
stream communities and ecosystem functions. Ecological Applications				
26(7), 2116-2129				
Mwedzi T., Zimunya T.G., Bere T., Tarakini T., Mangadze T. (2017).	Deposited	<2 mm for	Zimbabwe	Observational
Disentangling and ranking the influence of multiple stressors on	and	deposited		
macroinvertebrate communities in a tropical river system. International	suspended	sediment		
Review of Hydrobiology 102(5-6), 103-113				
Navel, S., Mermillod-Blondin, F., Montuelle, B., Chauvet, E., Simon, L.,	Deposited	<1 mm	France	Experimental
Piscart, C., Marmonier, P. (2010). Interactions between fauna and				
sediment control the breakdown of plant matter in river sediments.				
Freshwater Biology 55(4), 753-766				
Niyogi, D.K., Koren, M., Arbuckle, C.J., Townsend, C.R. (2007).	Deposited	Not recorded	New	Observational
Longitudinal changes in biota along four New Zealand streams:	and		Zealand	
Declines and improvements in stream health related to land use. New	suspended			
Zealand Journal of Marine and Freshwater Research 41(1), 63-75				
Niyogi, D.K., Koren, M., Arbuckle, C.J., Townsend, C.R. (2007). Stream	Deposited	<2 mm	New	Observational
communities along a catchment land-use gradient: Subsidy-stress			Zealand	
responses to pastoral development. Environmental Management 39(2),				
63-75				

Full reference	Sediment	Sediment size	Country	Study type
	fraction			
Nuttall, P.M., Bielby, G.H. (1973). The effect of China-clay wastes on	Suspended	Not recorded	UK	Observational
stream invertebrates. Environmental Pollution (1970) 5(2), 77-86				
O'Callaghan, P., Jocqué, M., Kelly-Quinn, M. (2015). Nutrient- and	Deposited	<2 mm	Honduras	Experimental
sediment-induced macroinvertebrate drift in Honduran cloud forest				
streams. Hydrobiologia 758(1), 75-86				
Osmundson, D.B., Ryel, R.J., Lamarra, V.L., Pitlick, J. (2002). Flow-	Deposited	<2 mm	USA	Observational
sediment-biota relations: Implications for river regulation effects on				
native fish abundance. Ecological Applications 12(6), 1719-1739				
Peeters, E.T.H.M., Brugmans, B.T.M.J., Beijer, J.A.J., Franken, R.J.M.	Deposited	<1 mm	Netherland	Experimental
(2006). Effect of silt, water and periphyton quality on survival and			S	
growth of the mayfly Heptagenia sulphurea. Aquatic Ecology 40(3),				
373-380				
Pereda O., Arroita M., Aristi I., Flores L., Larrañaga A., Elosegi A.	Deposited	<1 mm	Spain	Experimental
(2017). Effects of aeration, sediment grain size and burial on stream				
litter breakdown and consumer performance: A microcosm study.				
Marine and Freshwater Research 68(12), 2266-2274				
Phillips, I.D., Davies, JM., Bowman, M.F., Chivers, D.P. (2016).	Suspended	<63 µm	Canada	Observational
Macroinvertebrate communities in a Northern Great Plains river are				
strongly shaped by naturally occurring suspended sediments:				
Implications for ecosystem health assessment. Freshwater Science				
35(4), 1354-1364				
Piggott, J.J., Lange, K., Townsend, C.R., Matthaei, C.D. (2012).	Deposited	0.2 mm	New	Experimental
Multiple Stressors in Agricultural Streams: A Mesocosm Study of			Zealand	
Interactions among Raised Water Temperature, Sediment Addition and				
Nutrient Enrichment. PLoS ONE 7(11), e49873				

Full reference	Sediment	Sediment size	Country	Study type
	fraction			
Piggott, J.J., Townsend, C.R., Matthaei, C.D. (2015). Climate warming	Deposited	<2 mm	New	Experimental
and agricultural stressors interact to determine stream			Zealand	
macroinvertebrate community dynamics. Global Change Biology 21(5),				
1887-1906				
Pollard, A.I., Yuan, L.L. (2010). Assessing the consistency of response	Deposited	<2 mm	USA	Observational
metrics of the invertebrate benthos: A comparison of trait- and identity-				
based measures. Freshwater Biology 55(7), 1420-1429				
Quadroni, S., Brignoli, M.L., Crosa, G., Gentili, G., Salmaso, F.,	Deposited	Not recorded	Italy	Observational
Zaccara, S., Espa, P. (2016). Effects of sediment flushing from a small	and			
Alpine reservoir on downstream aquatic fauna. Ecohydrology 9(7),	suspended			
1276-1288				
Quist, M.C., Schultz, R.D. (2014). Effects of management legacies on	Deposited	Modified	USA	Observational
stream fish and aquatic benthic macroinvertebrate assemblages.		Wentworth -		
Environmental Management 54(3), 449-464		assume <2 mm		
Rabení, C.F., Doisy, K.E., Zweig, L.D. (2005). Stream invertebrate	Deposited	<2 mm	USA	Observational
community functional responses to deposited sediment. Aquatic				
Sciences 67(4), 395-402				
Ramezani, J., Rennebeck, L., Closs, G.P., Matthaei, C.D. (2014).	Deposited	Mean particle	New	Experimental
Effects of fine sediment addition and removal on stream invertebrates		size 0.2 mm	Zealand	
and fish: A reach-scale experiment. Freshwater Biology 59(12), 2584-				
2604				

Full reference	Sediment	Sediment size	Country	Study type
	fraction			
Reid, D.J., Chiaroni, L.D., Hewitt, J.E., Lohrer, D.M., Matthaei, C.D.,	Deposited	ranges from	New	Experimental
Phillips, N.R., Scarsbrook, M.R., Smith, B.J., Thrush, S.F., Townsend,		small gravel to	Zealand	
C.R., Van Houte-Howes, K.S.S., Wright-Stow, A.E. (2011).		clay		
Sedimentation effects on the benthos of streams and estuaries: A				
cross-ecosystem comparison. Marine and Freshwater Research 62(10),				
1201-1213				
Rosenberg, D.M., Wiens, A.P. (1978). Effects of sediment addition on	Suspended	98% was <2	Canada	Experimental
macrobenthic invertebrates in a Northern Canadian River. Water		mm		
Research 12(10), 753-763				
Sanpera-Calbet, I., Chauvet, E., Richardson, J.S. (2012). Fine sediment	Suspended	Kaolin - <45	France	Experimental
on leaves: Shredder removal of sediment does not enhance fungal		μm		
colonisation. Aquatic Sciences 74(3), 527-538				
Scott, Susanna E. Yixin Zhang (2012). Contrasting Effects of Sand	Deposited	sand	USA	Experimental
Burial and Exposure on Invertebrate Colonization of Leaves. American				
Midland Naturalist 167(1), 68-79				
Shaw, E.A., Richardson, J.S. (2001). Direct and indirect effects of	Suspended	< 425 µm	Canada	Experimental
sediment pulse duration on stream invertebrate assemblages and				
rainbow trout (Oncorhynchus mykiss) growth and survival. Canadian				
Journal of Fisheries and Aquatic Sciences 58(11), 2213-2221				
Strand, R.M., Merritt, R.W. (1997). Effects of episodic sedimentation on	Suspended	<0.6 mm	USA	Experimental
the net-spinning caddisflies Hydropsyche betteni and Ceratopsyche				
sparna (Trichoptera: Hydropsychidae). Environmental Pollution 98(1),				
129-134				

Full reference	Sediment	Sediment size	Country	Study type
	fraction			
Suren, A.M., Jowett, I.G. (2001). Effects of deposited sediment on	Deposited	47% silt and	New	Experimental
invertebrate drift: An experimental study. New Zealand Journal of	and	clay (<63 µm),	Zealand	
Marine and Freshwater Research 35(4), 725-737	suspended	20 % fine sand		
		(63 µm - 0.125		
		mm), 20%		
		medium sand		
		(0.125 mm -		
		0.5 mm), 9%		
		coarse sand		
		(0.5 - 2mm),		
		and 4% >2 mm		
Suren, A.M., Martin, M.L., Smith, B.J. (2005). Short-term effects of high	Suspended	Describes 'clay'	New	Experimental
suspended sediments on six common New Zealand stream		but also that	Zealand	
invertebrates. Hydrobiologia 548(1), 67-74		particles >0.05		
		mm were		
		allowed to		
		settle.		
Sutherland, A.B., Culp, J.M., Benoy, G.A. (2012). Evaluation of	Deposited	<2 mm	Canada	Observational
deposited sediment and macroinvertebrate metrics used to quantify				
biological response to excessive sedimentation in agricultural streams.				
Environmental Management 50(1), 50-63				
Suttle, K.B., Power, M.E., Levine, J.M., McNeely, C. (2004). How fine	Deposited	<2 mm	USA	Experimental
sediment in riverbeds impairs growth and survival of juvenile salmonids.				
Ecological Applications 14(4), 969-974				

Full reference	Sediment	Sediment size	Country	Study type
Taulhas W// Nistah O.T. Drawn D. Damalwishnan D. Tamalwas	fraction	0.000		Fun enime entel
Taulbee, W.K., Nietch, C.I., Brown, D., Ramakrishnan, B., Tompkins,	Deposited	<2 mm	USA	Experimental
W.J. (2009). Ecosystem consequences of contrasting flow regimes in an				
Descurres Association 45(4), 007,007				
Resources Association 45(4), 907-927				
Townsend, C.R., Uhlmann, S.S., Matthaei, C.D. (2008). Individual and	Deposited	<0.2 mm for	New	Both
combined responses of stream ecosystems to multiple stressors.		experiment	Zealand	
Journal of Applied Ecology 45(6), 1810-1819		and <2 mm for		
		field survey		
Turley, M.D., Bilotta, G.S., Chadd, R.P., Extence, C.A., Brazier, R.E.,	Deposited	<2 mm	UK	Observational
Burnside, N.G., Pickwell, A.G.G. (2016). A sediment-specific family-				
level biomonitoring tool to identify the impacts of fine sediment in				
temperate rivers and streams. Ecological Indicators 70(), 151-165				
Turley, M.D., Bilotta, G.S., Extence, C.A., Brazier, R.E. (2014).	Deposited	<2 mm	UK	Observational
Evaluation of a fine sediment biomonitoring tool across a wide range of	and			
temperate rivers and streams. Freshwater Biology 59(11), 2268-2277	suspended	_		
Turley, M.D., Bilotta, G.S., Krueger, T., Brazier, R.E., Extence, C.A.	Deposited	<2 mm	UK	Observational
(2015). Developing an improved biomonitoring tool for fine sediment:				
Combining expert knowledge and empirical data. Ecological Indicators				
54(), 82-86				
Turunen J., Louhi P., Mykrä H., Aroviita J., Putkonen E., Huusko A.,	Deposited	10 litres of fine	Finland	Experimental
Muotka T. (2018). Combined effects of local habitat, anthropogenic		sand at <2 mm		
stress, and dispersal on stream ecosystems: a mesocosm experiment.		and 10 litres of		
Ecological Applications 28(6), 1606-1615		coarse 2-3 mm		

Full reference	Sediment	Sediment size	Country	Study type
	fraction			
Vadher, A.N., Stubbington, R., Wood, P.J. (2015). Fine sediment	Deposited	1-2 mm	UK	Experimental
reduces vertical migrations of Gammarus pulex (Crustacea:				
Amphipoda) in response to surface water loss. Hydrobiologia 753(1),				
61-71				
Wagenhoff, A., Townsend, C.R., Phillips, N., Matthaei, C.D. (2011).	Deposited	<2 mm	New	Observational
Subsidy-stress and multiple-stressor effects along gradients of			Zealand	
deposited fine sediment and dissolved nutrients in a regional set of				
streams and rivers. Freshwater Biology 56(9), 1916-1936				
Wagenhoff, Annika Townsend, Colin R. Matthaei, Christoph D. (2012).	Deposited	<2 mm	New	Experimental
Macroinvertebrate responses along broad stressor gradients of			Zealand	
deposited fine sediment and dissolved nutrients: a stream mesocosm				
experiment. Journal of Applied Ecology 49(4), 892-902				
Wolmarans, C.T., Kemp, M., de Kock, K.N., Wepener, V. (2017). The	Deposited	<2000 µm	South	Observational
possible association between selected sediment characteristics and the			Africa	
occurrence of benthic macroinvertebrates in a minimally affected river in				
South Africa. Chemistry and Ecology 33(1), 18-33				
Zhu B., Smith D.S., Benaquista A.P., Rossi D.M., Kadapuram B.M., Yu	Suspended	Not recorded	USA	Observational
M.L., Partlow A.S., Burtch N.R. (2018). Water quality impacts of small-				
scale hydromodification in an urban stream in Connecticut, USA.				
Ecological Processes 7(11), doi:10.1186/s13717-018-0122-z				
Zweig, L.D., Rabeni, C.F. (2001). Biomonitoring for deposited sediment	Deposited	<2 mm	USA	Observational
using benthic invertebrates: A test on 4 Missouri streams. Journal of the				
North American Benthological Society 20(4), 643-657				

### Appendix 1.3 Number of articles per country included in evidence review

Country	Number	Country	Number	Country	Number
Austria	1	Phillipines	1	Spain	4
Bolivia	1	Taiwan	1	France	6
China	1	Zimbabwe	1	Ireland	6
Denmark	1	Croatia	2	Italy	6
Honduras	1	Finland	2	Australia	8
Japan	1	South Africa	2	Canada	12
Korea	1	Germany	3	UK	15
		Paraguay-			
Netherlands	1	Argentina	3	New Zealand	19
New					
Caledonia	1	Brazil	4	USA	27

Table A1.2 – Number of articles per country

#### Appendix 1.4 Frequency of responses recorded from evidence review

Response	Positive	Negative	Mixed	No	Total
category			response	effect	frequency
Population subset	47	177	37	176	437
Traits or trait-based					
index	59	120	22	118	319
Abundance/density	5	31	8	17	61
Richness	1	33	7	25	66
Biomonitoring index	6	99	14	20	139
Other	23	99	24	94	240
Diversity index	0	10	4	17	31
TOTALS	141	569	116	467	1293

Table A1.3 – Frequency of responses under each category

### Appendix 1.5 Residual tables from Chi-squared tests

Table A1.4 – Residuals from the Chi-squared test between the particle size and the detectable response using weight of evidence.

	Significant effect	Mixed response/no effect
0.25 mm	-1.811	2.131
>0.25 - 0.5 mm	-0.856	1.007
>0.5 - 1 mm	1.09	-1.282
>1 - 2 mm	2.511	-2.954
≥2 mm	-3.03	3.565
Not specified	-0.87	1.023

Table A1.5 – Residuals from the Chi-squared test between the taxonomic level and detectable effect where the subset of the population was recorded as an abundance.

	Significant effect	Mixed response/no effect
Phylum/subphylum	0.049	-0.05
Class	0.82	-0.824
Subclass	-1.228	1.233
Order	-1.075	1.079
Family	-0.7	0.703
Subfamily	1.404	-1.409
Genus	1.088	-1.092
Species	-0.351	0.353

Table A1.6 – Residuals from the Chi-squared test between the taxonomic level and detectable effect where the subset of the population was recorded as relative abundance

	Significant effect	Mixed response/no effect
Phylum/subphylum	0.589	-0.456
Class	0.809	-0.626
Subclass	0.589	-0.456
Order	1.714	-1.327
Family	-3.274	2.536
Subfamily	0.923	-0.715
Genus	0.807	-0.625
Species	1.652	-1.279

Table A1.7 – Residuals from the Chi-squared test between traditional metrics and the detectable effect

	Mixed response/no effect	Negative	Positive
Abundance/density	-1.355	0.567	2.892
Richness	0.352	0.141	-1.773
Diversity	1.629	-1.185	-1.696

Table A1.8 – Residuals from the Chi-squared test between functional feeding group and the detectable effect where the functional feeding group was recorded as an abundance

	Mixed response/no effect	Negative	Positive
Collector	2.297	-1.361	-0.766
Filterer	-1.376	0.655	0.901
Other	1.067	-1.377	1.703
Predator	0.941	-0.301	-1.022
Scraper	1.01	-0.918	0.546
Shredder	-2.034	1.731	-0.773

Table A 1.9 – Residual table from the Chi-squared test between functional feeding group and the detectable effect where the functional feeding group was recorded as relative abundance

	Mixed response/no effect	Negative	Positive
Collector	-1.023	-0.734	2.333
Filterer	-1.221	0.346	2.037
Other	0.153	-0.125	-0.205
Predator	0.64	-0.288	-1.004
Scraper	0.737	0.201	-1.48
Shredder	0.842	0.379	-1.782

Table A1.10 – Residual table from the Chi-squared test between the trait group 'mode of locomotion' and 'relationship to substrate' and the effect where the trait modality was recorded as abundance

	Mixed response/no effect	Negative	Positive
Attached	0.199	0.289	-0.631
Burrower	0.117	-2.512	3.794
Climber	1.225	-0.324	-0.614
Clinger	-1.721	2.225	-1.886
Crawler	1.766	-0.946	-0.142
Flier	1.572	-0.653	-0.42
Interstitial	-0.829	-0.347	1.295
Sprawler	-0.56	0.724	-0.614
Surface swimmer	1.572	-0.653	-0.42
Swimmer	0.132	0.77	-1.316

Table A1.11 – Residual table from the Chi-squared test between the trait group 'mode of locomotion' and 'relationship to substrate' and the effect where the trait modality was recorded as relative abundance

	Mixed response/no effect	Negative	Positive
Burrower	-2.12	-2.258	3.829
Climber	2.196	-1.34	-1.461
Clinger	-0.244	2.687	-1.566
Crawler	0.829	1.886	-2.179
Depositional	1.252	-0.63	-0.925
Erosional	-0.999	2.944	-0.925
Sprawler	0.914	-0.46	-0.675
Swimmer	0.64	-0.025	-0.675

Table A1.12 – Residual table from the Chi-squared test between biomonitoring indices and the detectable effects

	Mixed response/no effect	Negative	Positive
General index	1.14	-0.84	0.28
Sediment specific	-2.898	2.136	-0.712
Stressor specific	-1.962	1.446	-0.482

## Appendix 1.6 Diagnostic plots of linear model determining significant predictors of evidence quality.



Figure A1.1– Diagnostic plots from refined model (EcoEvidence score is the model response)

## Appendix 2



Appendix 2.1 SEM images of *Ecdyonurus venosus* gills from pilot study

Figure A2.1 – Scanning Electron Microscopy images from *E. venosus* individuals observed immediately after sampling from a local site (a,c) and those which had undergone sediment exposure as part of a pilot study (b,d).

# Appendix 2.2 Turbidity, suspended sediment concentrations and velocity for each trial

Table A2.1 - Target turbidity, mean turbidity (from 1 s resolution sonde data), mean suspended sediment concentrations and mean velocity ( $\pm$  1 standard deviation) for each experimental trial.

Trial	Target turbidity (NTU)	Mean turbidity (NTU)	Mean suspended sediment concentration (mg I <sup>-1</sup> )	Mean velocity (m s <sup>-1</sup> )
1	< 2.5	1.29 (0.12)	3.82 (1.32)	0.19 (0.003)
2	< 2.5	2.76 (0.41)	3.19 (3.19)	0.41 (0.01)
3	100	101.27 (5.61)	81.02 (7.94)	0.19 (0.004)
4	100	101.94 (4.38)	86.31 (6.55)	0.34 (0.01)
5	400	401 (11.68)	368.52 (42.05)	0.19 (0.01)
6	400	399.49 (8.90)	439.97 (88.39)	0.35 (0.01)



Appendix 2.3 Particle size distribution of the experimental sediment

Figure A2.2 - Particle size distribution curve of the sediment aggregate mix added to the recirculating flume system during the experiments. The particle size distribution was calculated using laser particle size analysis and is an average of two samples from each of two duplicate runs.



### Appendix 2.4 Microscopy images of gills from each experimental run

Figure A2.3 – Examples of images of slide mounts of invertebrate gills for each of *Ecdyonurus venosus* (10 X magnification), *Ephemera danica* (10 X magnification) and *Hydropscyhe siltalai* (20 X magnification) after exposure two controls and four treatments of varying SSC and flow velocity. Control (1) = 3.5 mg l<sup>-1</sup> at 0.19 m s<sup>-1</sup>, control (2) = 3.5 mg l<sup>-1</sup> at 0.37 m s<sup>-1</sup>, treatment (3) = 83.7 mg l<sup>-1</sup> at 0.19 m s<sup>-1</sup>, treatment (4) = 83.7 mg l<sup>-1</sup> at 0.37 m s<sup>-1</sup>, treatment (5) = 404.0 mg l<sup>-1</sup> at 0.19 m s<sup>-1</sup> and treatment (6) = 404.0 mg l<sup>-1</sup> at 0.37 m s<sup>-1</sup>. The three gill types structures have been labelled on an image of each species.

### Appendix 2.5 Three-way ANOVA results

Term	Df	SS	Estimate	F	p
Species	2	1.406	0.703	29.499	<0.001*
Sediment	2	1.021	0.510	21.406	<0.001*
Velocity	1	0.047	0.047	1.964	0.165
Species:Sediment	4	0.827	0.207	8.670	<0.001*
Species:Velocity	2	0.270	0.135	5.671	0.005*
Sediment:Velocity	2	0.046	0.023	0.956	0.389
Species:Sediment:Velocity	4	0.537	0.134	5.627	<0.001*
Residuals	82	1.955	0.024		

Table A2.2 - Summary results from the three-way ANOVA. \*Denotes a significant term (p <0.05).

### Appendix 2.6 Model selection results

Table A2.3 - Summary results from the model selection procedure. \*Denotes that the model including the interaction is a significantly better fit than the simpler model (p < 0.05).

Model	Res. Df	RSS	Df	SS	F	p	AICc
E. venosus							
Sediment +	29	0.443					-36.386
Velocity							
Sediment *	27	0.398	2	0.045	1.535	0.234	-33.682
Velocity							
E. danica							
Sediment +	27	0.977					-9.203
Velocity							
Sediment *	25	0.880	2	0.097	1.373	0.272	-8.433
Velocity							
H. siltalai							
Sediment +	32	1.117					-12.867
Velocity							
Sediment *	30	0.677	2	0.440	9.759	<0.001*	-23.908
Velocity							

### **Appendix 3**

### Appendix 3.1 Scoring system and formulas for national fine sedimentspecific biomonitoring indices

Table A3.1 - Fine sediment sensitivity ratings and abundance weightings for the PSI index (from Extence et al. 2011)

Group	Fine Sediment Sensitivity	Log abundance			
	Rating (FSSR)	1-9 10-99 100-		100-	1000+
				999	
A	Highly sensitive	2	3	4	5
В	Moderately sensitive	1	2	3	4
С	Moderately insensitive	1	2	3	4
D	Highly Insensitive	2	3	4	5

$$PSI = \frac{\sum \text{Scores for Sediment Sensitivity Groups A \& B}}{\sum \text{Scores for all Sediment Sensitivity Groups A; B; C \& D}} X 100$$

Equation A3.1 - PSI index equation where A, B, C and D are the Fine Sediment Sensitivity Ratings corresponding with Table 3 (Extence et al. 2013).

$$E - PSI = \frac{\sum(logA_{sens} \times W)}{\sum(logA_{all} \times W)} \times 100$$

Equation A3.2 - EPSI index equation where  $logA_{sens}$  is the log abundance categories for sensitive species and their corresponding weighting W and  $logA_{all}$  is the log abundance categories for all species.

Table A3.2 – shows the eigenvalues for the partial canonical correspondence analysis (pCCA), from Murphy et al. (2015).

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*CoFSIsp* = 0.349oFSIsp + 0.569ToFSIsp

Equation A3.3 – CoFSI equation where oFSI is the organic fine sediment index score and ToFSI is the total fine grain sediment index score.



Appendix 3.2 Maps outlining screening process for field site selection

Figure A3.1 – Maps showing the reduction of suitable sites at each screening phase starting with sites passing water chemistry requirements (a), sites in lowland end groups (b), sites without re-sectioning or capital works (d), sites matched by season/year (d), all active gauging stations in England, Scotland and Wales € and sites within 2km of active gauging stations (f).

### Appendix 3.3 Example of an Environment Agency site card



Site Identification							
Area Name	South East						
Region Name	Thames						
Site Id	47269						
Sample Point Refs	BTH35614						
Site Name	TILLINGBOURNE - EAST SHALFORD LANE						
Water Body	Tillingbourne						

Site Address / Location					
Routing Grid Reference	TQ0120047400				
Site Grid Reference	TQ0119347418				
Local Directions	Drive in to East Shalford Lane and follow until the bridge over the Tillingbourne. You can also park a wee bit uphill from the bridge, where a footpath joins the road. Access to the river is very easy upstream of the bridge on the true right bank.				

Site Risk Assessment Information						
Mobile Signal Strength At Site	Good					
Overall Risk Assessment Level	Low					
Number of Samplers Required	BTH35614: 1	3				

File Last Updated

12/09/2016

Detailed Information						
Additional Site Instructions	There is often a large quantity of grass clippings and other garden waste dumped on the left bank, which can fall in to the river. £					
Additional Sampling Time Allowed (mins)	Sampling Point BTH35614: 5 This item has been removed due to third party copyright. The unabridged version of the thesis can be viewed at the Lanchester library, Coventry University					
Sampling Point BTH35614. Image: tillingboune at east shallford lane	Sampling Point BTH35614. Image: P4190225. Park here					

Figure A3.2 – Example of a site card obtained from the Environment Agency used to determine location of sampling area

	Appendix	3.4 Field	sheet
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Morwenna Mckenzie												
PhD Coventry												
Field sampling sheet												
SITE ID/Waterbody ID				Eastings			5	Northings				
RIVER				DATE			TIME			SAMPL	ER	
	Τ		SITE DIA	GRAM A	AND ECO	LOGI	CAL CO	MMENT	rs			
Width of river	D	epth of river	STE DAGAAN AND ECODOSICAL COMMENTS									
1	1											
-	2											
2	_											
	3_											
AVERAGE	AV	SAMPLING										
WIDTH (m)	DEI	2TH (cm)										
			% pool		%							
SUBSTRATE %		Time allocation	% glide		%							
BEDROCK		IOI KICK HEL	% riffle		%							
BOULDERS		Pool secs	% other	(	%)							
COBBLES		Glide secs	DETH	RITUS S	EWAGE	BED	STABIL	ITY O	VERL	AYING	OVER	LAYING
PEBBLES		D:00-		1	LITTER			3	SILT C	OVER	SILTD	ENSITY
GRAVEL		KIIIIe Secs	NONE	0	0	SOLI	D	0 N	ONE	0	TRACE	0
SAND		Other secs	LOCAL	0	0	STAR	SLE	0 5	OCAL 30%	0	THIN	0
		l minute hand	<30%			LOO	OF	ő w	VIDESP	READ O	THICK	- 0
SILT		search o	WIDESPRE 30-60%	ead O	0	200	-	0 30	0-60%		MASSI	VE 0
CLAY			EXTENSIV	τo	0	DAN	GEROUS	0 6	XTENS 0-100%	IVE O		
	Ť	ODOUR	TURBIDIT	¥	SHADE			FLOW	v	1	INFLUEN	CES
RUBBISH O		NONE O	CLEAR	0	HEAVY	>50%	0	DRY		。	SALINE	0
OIL FILM O		SLIGHT 0	SLIGHT	õ	LIGHT	-25%	ŏ	NO FL	low	°	DREDGIN	ic 0
OIL DEPOSIT O		STRONG 0	MOD	0	MOD	25-50	96 O	NORM	(AL	õ	WEED CI	то
ERODING U			HIGH	0	NONE		0	HIGH	F	ŝ		
SEWAGE FUNGUS ON STONES FILAMENTOUS NON-FILAMENTOUS												
HABITAT			ABOVE (S	BELOW	OCHRE	2	LGAE		ALGAE	MAC	ROPHYTE	MOSS
TORRENT	~	NONE	0	٥	0		0		۰		0	•
RIFFLE	~	WIDESPREAD 30-0	0%6 0	0	°		0		0		0	°
RUN	0	EXTENSIVE 60-100	% o	۰	۰		۰		۰		0	•
GLIDE	0	PERCENTAGE	-		_				_			
SLACK	0	NOT RECORDED	•	•	•		•		•		•	°
DITCH	0	DENSITY			0							
WATERFALL	0	THIN	õ	õ	õ		õ		õ			
CASCADE	0	THICK MASSIVE	°	°	0		0		0			
RAPID	0	LAND USE	PRM SEC	2			PRM SE	ic B	SANK S	TRUCTU	URE	
PONDED REACH	0	BROADLEAF WOO	вО Оз	CRUB		0	0 0				PRM	SEC
MARG DEAD		CONIFEROUS WOO	оо от	ALL HER	ES/RANK	0	0 0	Р	REDO	dinantly	¥	
WATER	0	OPEN WATER	0 011	ILLED L	AND	(	0 0	1	BARE G	ROUND	0	0
EXPOSED		SUE/URBAN	0 013	(PROVE)	D PASTUR	E (	0 0	1	DOMN	EGTYPE	0	0
BEDROCK	0	ROCK & SCREE	0 0 0	OUGH P/	STURE	(	0 0					-
MATURE ISLAND	0	ORCHARD	0 015	DUSTRI	AL		0	2	-3 DOM	UNANT	PES 0	
UNVEG MID BAR	0	MOOBLAND	0 00 0 000	ARM BUI	LDINGS			``	LOLIA	103 11		v
VEG MID BAR	0	BADKS CARDENS		OAD/RAI	LWAY		, ,	>	4 DOM	INANT		_
INVERSING SINC DAD	0	PARAST GARDENS						v	EGETA	TION TY	res O	0

Figure A3.3 – The field sheet completed at each sampling occasion
### Appendix 3.5 Taxonomic identification resolution

Table A3.3 – Taxonomic resolution that each phylum is identified to as part of Environment Agency protocols (from Environment Agency 2014b) This item has been removed due to third party copyright. The unabridged version of the thesis can be viewed at the Lanchester library, Coventry University





Figure A3.4 – PCA plot for hydrological regime metrics



Figure A3.5 – Contribution of each flow regime metric to the first two principal components.



Figure A3.6 – Correlation matrix of flow regime metric. Colour ramp indicates strength and direction (positive or negative) of correlation coefficient.



Figure A3.7 – PCA plot of antecedent flow metrics.



Figure A3.8 – Contribution of each antecedent flow metric to principal components 1 and 2.

Correlation matrix for antecedent flow metrics is not shown due to the large number of variables.

### Appendix 3.7 List of predictors for linear modelling (sediment metrics)

Table A3.4 – List of total environmental predictors and those retained after total reducing variance inflation factor.

Original predictors		Predictors retained after variance inflation factor
Total surface sediment, Organic surface sediment, Inorganic surface sediment, Total subsurface sediment, Total organic sediment, Total inorganic sediment, Visual fines, Width, Depth, Filamentous algae, Macrophyte, Altitude, Slope, Discharge, Distance from source, Background SSC, Coarse bed matrix, Erosional flow, Depositional flow, Stream power, Q20, Q10, DFQ95MEAN, DFMEDMAX, DAY90MAX, Q1, Q1090DF, DAY7MAX, Q95pre6m, Q95preSum, MIN7d, Q20preSpr, Q50pre30d, MDFpreSpr, MDF7d, Q20pre6m, Q20preWin, Q50preWin, Q50preAut, MINpreSpr, Q50preSum, Q10preSpr, Q20pre30d, MDFpreAut, Q10pre6m, MDFpreWin, Q10preAut, Q20pre7d	corvif() →	Width, Depth, Bedrock, Filamentous algae, Macrophyte, Altitude, Slope, Background SSC, Coarse bed matrix, Erosional flow, Stream power, Q1, Q1090DF, Q20pre6m, Q50preWin, Q50preSum, Q20pre7d

# Appendix 3.8 List of predictors for linear modelling (biomonitoring indices)

Table A3.5 – List of total environmental predictors and those retained after total reducing variance inflation factor.

Original predictors		Predictors retained after
		variance inflation factor
Visual fines reach, Total surface, Organic surface, Inorganic surface, Total subsurface, Total organic, Total inorganic, Visual fines patch, Background SSC, RIVPACS end group, Super end group, ASL, Width, Depth, Bedrock, Detritus, Bed stability, Shade, Flow, Filamentous algae, Macrophyte, Altitude, Slope, Discharge, Distance from source, Coarse bed matrix, Erosional flow, Depositional flow, Q20, Q10, DFQ95MEAN, DFMEDMAX, DAY90MAX, Q1, Q1090DF, DAY7MAX, Q95pre6m, Q95preSum, MIN7d, Q20preSpr, Q50pre30d, MDFpreSpr, MDF7d, Q20pre6m, Q20preWin, Q50preWin, Q50preAut, MINpreSpr, Q50preSum, Q10preSpr, Q20pre30d, MDFpreAut, Q10pre6m, MDFpreWin, Q10preAut, Q50preSyr, Q10preAut, Q50preSyr, Q10preAut, Q20preAut, Q20preZd	corvif() →	variance inflation factor Organic surface, Total inorganic, Background SSC, Q1090DF, Q20pre6m, Q50preWin, Q50preSum, Q20pre7d, End group, ASL, Depth, Bedrock, Detritus, Filamentous algae, Macrophyte, Altitude, Slope, Distance from source, Coarse bed matrix, Stream power

### Appendix 3.9 Model selection process (biomonitoring indices)

Table A3.6 – Model selection process showing Akaike's Information Criterion (AIC) for the model combinations for each metric of fine sediment. Optimal model denoted with an asterisk.

Model	(environmental variables as fixed predictors)	(environmental variables as fixed predictors) + season	(environmental variables as fixed predictors) + season + (1 season)	(environmental variables as fixed predictors) + (1 season)
PSI	342.202	339.970*	344.102	341.970
EPSI	334.653*	335.317	336.653	337.317
EPSImixed	335.551*	335.945	337.551	337.945
CoFSI	6.138*	4.696	8.138	6.696
oFSI	44.662	34.609*	41.440	36.609
ToFSI	22.414*	24.412	24.414	26.412
WHPT ASPT	83.735	79.400*	84.629	81.400
WHPT NTAXA	261.159*	262.974	263.159	264.974
LIFE	62.959	58.037*	63.493	60.037
EPT	322.722	315.731*	321.838	317.730
FRic	-332.645	-335.502*	-330.976	-333.502
FDis	131.147*	133.144	133.147	135.144

### Appendix 3.10 Taxa included in TITAN analysis

Table A3.7 – List of taxa which occurred at three or more sites (and used in TITAN analysis)

Agapetus sp	Ephemera sp	Mystacides azurea
Anabolia nervosa	Erpobdella octoculata	Mystacides longicornis
Ancylus fluviatilis	Erpobdellidae	Mystacides sp
Asellus aquaticus	Esolus parallelepipedus	Odontocerum albicorne
Athripsodes albifrons	Gammarus fossarum	Oligochaeta
	Gammaraus	
Athripsodes bilineatus	fossarum/pulex	Orectochilus villosus
		Orthocladiinae/Diamesinae
Athripsodes cinereus	Gammarus pulex	/ Prodiamesinae
Athripsodes sp	Gammarus sp	Ostracoda
Baetidae	Glossiphonia complanata	<i>Oulimnius</i> sp
Baetis rhodani	Glossiphonia heteroclita	Pericoma sp
Baetis scambus group	Boreobdella verrucata	Piscicola geometra
<i>Baetis</i> sp	Glossosomatidae	Pisidium sp
Beraeodes minutus	Goera pilosa	Podonominae
Bithynia tentaculata	Goeridae	Polycelis felina
		Polycentropus
Brachycentrus subnubilus	Habrophlebia fusca	flavomaculatus
Caenis luctuosa group	Halesus sp	Polycentropus kingi
Caenis rivulorum	Helobdella stagnalis	Polycentropus sp
Calopteryx splendens	Heptageniidae	Potamopyrgus jenkinsi
Calopteryx virgo	Hydracarina	Potamophylax group
Centroptilum luteolum	Hydraenidae	Proasellus meridianus
Ceratopogonidae	Hydropsyche instabilis	Radix balthica
Chaetopteryx villosa	Hydropsyche pellucidula	Rhithrogena sp
Chironomidae	Hydropsyche siltalai	Rhyacophila dorsalis
Chironomini	Hydropscyhe sp	Rhyacophila sp
Cladocera	Hydroptilidae	Riolus sp
Crangonyx pseudogracilis	Lepidostoma hirtum	Sericostoma personatum
Cyrnus trimaculatus	Leptophlebiidae	Serratella ignita
Dendrocoelum lacteum	Leuctra fusca	Sialis lutaria
Diptera	Euleuctra geniculata	Silo nigricornis
Dixa nebulosa	Leuctra nigra	Simuliidae
Drusus annulatus	Limnephilus lunatus	Sphaerium corneum
Dytiscidae	Limnephilidae	Tanypodinae
Ecdyonurus sp	Limnius volckmari	Tanytarsini
Elmis aenea	Limoniidae	Theodoxus fluviatilis
Empididae	<i>Lype</i> sp	Tipulidae
Ephemera danica		

Appendix 3.11 Scree plot for PCA of all environmental variables at each site



Figure A3.9 – A scree plot showing the contribution of each dimension to the total explained variance for PCA of all environmental variables for all sites

### Appendix 3.12 Sediment data for each field site

Table A3.8 -	Fine sediment results	of visual assessmer	nts and resuspe	ension sampling a	at each field site for each season.
1 4010 / 1010			no ana roodope	noion oampning c	

				Visual	Total	Organic	Inorganic	Total	Organic subsurface	Inorganic
Site	Patch	Season	Patch	(%)	m <sup>-2</sup> )	m <sup>-2</sup> )	m <sup>-2</sup> )	(g m <sup>-2</sup> )	(g m <sup>-2</sup> )	(g m <sup>-2</sup> )
177	Depositional	Autumn	1	95	558.12	80.67	470.83	740.58	108.10	625.86
177	Depositional	Autumn	2	80	384.31	68.03	307.47	392.64	55.07	328.76
177	Erosional	Autumn	1	80	667.26	90.62	568.04	1024.43	132.31	883.52
177	Erosional	Autumn	2	70	111.83	10.74	91.83	463.16	62.46	391.45
177	Depositional	Spring	1	100	695.17	82.47	595.31	2727.13	285.43	2424.30
177	Depositional	Spring	2	75	260.68	10.13	239.02	819.26	73.05	734.68
177	Erosional	Spring	1	70	478.54	34.15	437.78	1997.38	174.27	1816.51
177	Erosional	Spring	2	70	192.49	31.05	152.74	192.49	31.05	152.74
1313	Depositional	Autumn	2	0	0.08	0.00	2.72	0.00	0.00	0.00
1313	Depositional	Autumn	1	0	2.75	0.00	4.13	24.68	0.00	22.45
1313	Erosional	Autumn	2	2	0.00	0.00	0.00	0.36	0.00	0.12
1313	Erosional	Autumn	1	0	0.79	0.00	0.73	0.00	0.00	0.40
1313	Depositional	Spring	1	0	492.26	131.76	357.01	338.10	84.44	250.18
1313	Depositional	Spring	2	0	20.97	4.26	11.78	52.79	7.79	40.06
1313	Erosional	Spring	1	0	3.69	0.00	0.00	7.62	0.00	2.40
1313	Erosional	Spring	2	0	1.11	0.00	0.00	19.25	0.00	12.16
7694	Depositional	Autumn	2	25	111.34	29.78	88.01	243.96	45.51	204.90
7694	Depositional	Autumn	1	0	165.89	32.04	143.29	451.35	61.43	399.35
7694	Erosional	Autumn	2	45	1.07	1.96	1.72	32.45	4.47	30.57
7694	Erosional	Autumn	1	55	19.89	7.09	17.78	23.58	7.61	20.94
7694	Depositional	Spring	1	10	47.14	0.00	32.22	145.55	0.00	122.12

				Visual	Total	Organic	Inorganic	Total	Organic	Inorganic
				fines	surface (g	surface (g	surface (g	subsurface	subsurface	subsurface
Site	Patch	Season	Patch	(%)	m <sup>-2</sup> )	m⁻²)	m⁻²)	(g m⁻²)	(g m⁻²)	(g m⁻²)
7694	Depositional	Spring	2	0	33.59	1.24	2.37	44.17	0.00	33.77
7694	Erosional	Spring	1	20	16.43	0.00	14.02	185.03	19.66	158.54
7694	Erosional	Spring	2	45	3.36	0.00	2.74	148.69	0.00	144.36
8614	Depositional	Autumn	2	95	1183.41	62.65	1101.23	1974.21	142.01	1812.67
8614	Depositional	Autumn	1	70	482.22	3.78	469.00	995.56	33.96	952.16
8614	Erosional	Autumn	2	30	5.41	0.00	5.78	218.86	16.55	199.26
8614	Erosional	Autumn	1	90	222.26	2.08	211.46	2081.67	63.55	2009.40
8614	Depositional	Spring	1	30	642.09	0.00	620.24	2149.55	756.55	1369.62
8614	Depositional	Spring	2	0	1010.40	37.08	954.32	1773.86	48.26	1706.61
8614	Erosional	Spring	1	0	3.57	0.00	3.08	49.13	0.84	43.67
8614	Erosional	Spring	2	0	7.48	0.00	7.53	58.69	0.00	52.31
9144	Depositional	Autumn	2	0	8.96	3.43	6.51	188.99	39.27	150.70
9144	Depositional	Autumn	1	30	52.33	19.66	34.98	229.11	46.36	185.07
9144	Erosional	Autumn	2	10	2.20	0.48	2.16	26.52	6.31	20.65
9144	Erosional	Autumn	1	10	1.43	1.50	0.89	34.55	6.99	28.53
9144	Depositional	Spring	1	0	14.68	0.00	6.38	156.72	8.42	133.58
9144	Depositional	Spring	2	0	0.00	0.00	0.00	16.25	0.00	12.33
9144	Erosional	Spring	1	0	0.18	0.00	0.00	8.85	0.00	6.79
9144	Erosional	Spring	2	0	1.05	0.00	0.62	25.84	2.87	20.05
10533	Depositional	Autumn	2	10	130.17	19.99	107.77	583.40	79.39	501.60
10533	Depositional	Autumn	1	10	0.00	0.00	0.00	31.28	0.00	25.16
10533	Erosional	Autumn	2	10	0.00	0.00	0.00	17.88	1.40	13.40
10533	Erosional	Autumn	1	5	0.00	0.00	0.00	0.99	0.00	0.00
10533	Depositional	Spring	1	10	10.87	0.00	10.11	19.29	0.00	15.81
10533	Depositional	Spring	2	40	16.11	0.00	14.47	163.88	9.42	148.14

				Visual	Total	Organic	Inorganic	Total	Organic	Inorganic
0:44	Datak	<b>C</b>	Detak	fines	surface (g	surface (g	surface (g	subsurface	subsurface	subsurface
Site	Patch	Season	Patch	(%)	m²)	m <sup>-</sup> *)	m <sup>-</sup> )	(g m²)	(g m²)	(g m²)
10533	Erosional	Spring	1	5	2.10	0.00	2.23	16.99	0.00	14.41
10533	Erosional	Spring	2	0	3.20	0.00	3.91	9.99	0.00	10.18
35479	Depositional	Autumn	2	10	20.64	9.43	13.96	294.21	45.66	251.30
35479	Depositional	Autumn	1	20	195.55	43.53	160.36	274.80	60.22	222.93
35479	Erosional	Autumn	2	20	24.90	8.20	18.48	117.51	27.79	91.50
35479	Erosional	Autumn	1	10	17.77	6.67	14.14	116.42	22.64	96.82
35479	Depositional	Spring	1	15	8.69	0.00	6.28	43.91	2.78	37.69
35479	Depositional	Spring	2	15	6.98	0.00	4.90	21.03	0.00	17.65
35479	Erosional	Spring	1	15	134.67	11.43	118.16	421.16	43.77	372.31
35479	Erosional	Spring	2	10	36.55	2.40	31.00	434.80	36.26	395.39
35614	Depositional	Autumn	2	95	162.96	1.85	159.71	698.81	14.06	683.34
35614	Depositional	Autumn	1	80	23.40	0.00	21.48	34.11	0.09	31.93
35614	Erosional	Autumn	2	60	289.53	19.54	267.69	221.65	7.56	211.79
35614	Erosional	Autumn	1	40	422.69	33.87	386.57	831.71	30.84	798.62
35614	Depositional	Spring	1	100	2011.78	68.19	1926.14	1706.23	208.26	1480.52
35614	Depositional	Spring	2	80	396.74	0.00	383.25	984.60	110.67	858.82
35614	Erosional	Spring	1	10	13.38	0.00	10.10	45.15	0.00	39.04
35614	Erosional	Spring	2	20	68.31	0.00	65.67	63.67	0.00	52.80
42051	Depositional	Autumn	2	100	128.47	19.38	109.09	291.47	33.72	257.75
42051	Depositional	Autumn	1	55	293.08	48.77	244.31	300.24	48.81	251.43
42051	Erosional	Autumn	2	10	7.04	0.83	6.21	5.20	0.58	4.62
42051	Erosional	Autumn	1	0	4.78	0.39	4.40	14.64	0.83	13.81
42051	Depositional	Spring	1	70	572.01	64.28	496.24	827.78	84.61	731.68
42051	Depositional	Spring	2	60	93.29	6.11	79.02	48.15	1.75	38.24
42051	Erosional	Spring	1	10	5.24	0.00	2.92	135.09	9.34	121.67

				Visual	Total	Organic	Inorganic	Total	Organic	Inorganic
		_		fines	surface (g	surface (g	surface (g	subsurface	subsurface	subsurface
Site	Patch	Season	Patch	(%)	m <sup>-2</sup> )	m <sup>-2</sup> )	_m <sup>-2</sup> )	(g m⁻²)	(g m⁻²)	(g m⁻²)
42051	Erosional	Spring	2	0	4.88	0.00	3.11	15.54	0.00	13.39
42744	Depositional	Autumn	2	33	103.17	13.18	91.26	227.91	28.80	200.37
42744	Depositional	Autumn	1	70	269.78	27.55	243.40	698.95	79.91	620.21
42744	Erosional	Autumn	2	5	0.44	0.26	0.39	0.00	0.21	0.00
42744	Erosional	Autumn	1	10	25.46	2.93	22.78	7.98	0.71	7.52
42744	Depositional	Spring	1	15	27.27	0.00	24.46	35.64	0.00	27.96
42744	Depositional	Spring	2	80	53.49	0.00	42.64	278.43	5.38	262.12
42744	Erosional	Spring	1	10	3.20	0.00	1.18	13.22	0.00	12.21
42744	Erosional	Spring	2	10	1.47	0.00	0.45	3.08	0.00	2.20
42794	Depositional	Autumn	2	70	694.56	174.17	519.48	1930.08	386.09	1543.08
42794	Depositional	Autumn	1	80	395.45	97.57	296.92	690.98	163.28	526.74
42794	Erosional	Autumn	2	40	60.44	6.93	53.14	606.95	71.34	535.24
42794	Erosional	Autumn	1	20	7.73	1.95	5.53	5.90	1.96	3.68
42794	Depositional	Spring	1	75	131.73	4.02	114.17	149.57	7.63	128.39
42794	Depositional	Spring	2	80	379.91	47.01	317.16	1095.18	107.48	971.96
42794	Erosional	Spring	1	0	12.68	0.00	11.14	18.57	0.03	15.16
42794	Erosional	Spring	2	70	44.54	0.81	35.07	506.06	40.89	456.51
43795	Depositional	Autumn	2	80	737.37	42.36	688.34	4176.42	236.22	3933.53
43795	Depositional	Autumn	1	10	150.88	6.46	137.19	475.93	31.14	437.55
43795	Erosional	Autumn	2	10	1166.33	97.73	1058.87	1525.80	119.47	1396.60
43795	Erosional	Autumn	1	70	164.57	7.62	150.66	231.19	11.83	213.07
43795	Depositional	Spring	1	75	247.55	0.00	239.82	852.60	17.79	820.05
43795	Depositional	Spring	2	80	200.71	0.00	194.61	395.46	0.00	392.89
43795	Erosional	Spring	1	60	34.81	0.00	39.87	60.52	0.00	61.40
43795	Erosional	Spring	2	70	148.69	0.00	144.36	625.35	0.00	833.14

				Visual	Total	Organic	Inorganic	Total	Organic	Inorganic
		_		fines	surface (g	surface (g	surface (g	subsurface	subsurface	subsurface
Site	Patch	Season	Patch	(%)	m <sup>-2</sup> )	m⁻²)	_m⁻²)	(g m⁻²)	(g m⁻²)	(g m⁻²)
49306	Depositional	Autumn	2	100	1122.94	190.44	947.70	708.29	113.63	609.86
49306	Depositional	Autumn	1	100	440.86	87.05	370.37	625.76	95.61	546.71
49306	Erosional	Autumn	2	100	483.45	64.06	432.82	1076.99	108.30	982.13
49306	Erosional	Autumn	1	100	2397.00	333.32	2076.27	5382.50	625.15	4769.93
49306	Depositional	Spring	1	90	218.41	0.00	182.36	621.27	0.00	585.75
49306	Depositional	Spring	2	95	194.68	0.00	166.87	249.40	0.00	212.37
49306	Erosional	Spring	1	25	342.38	31.48	281.68	2751.50	171.51	2550.77
49306	Erosional	Spring	2	80	286.16	0.00	258.00	2697.33	161.51	2497.48
54650	Depositional	Autumn	2	80	175.07	40.63	145.64	865.66	70.26	806.60
54650	Depositional	Autumn	1	80	259.34	68.06	194.26	1719.86	336.79	1386.05
54650	Erosional	Autumn	2	0	6.91	3.14	4.35	302.61	58.72	244.46
54650	Erosional	Autumn	1	2	3.32	1.90	2.58	280.74	36.86	245.04
54650	Depositional	Spring	1	10	5.71	0.00	6.08	145.27	16.16	125.59
54650	Depositional	Spring	2	40	42.88	5.66	34.09	444.32	80.83	360.35
54650	Erosional	Spring	1	5	18.57	0.55	16.55	109.42	11.81	96.14
54650	Erosional	Spring	2	0	19.07	0.00	17.73	187.51	17.30	168.60
65511	Depositional	Autumn	2	0	1.51	0.00	0.69	4.79	0.24	3.72
65511	Depositional	Autumn	1	0	3.78	1.04	1.82	4.20	0.20	3.08
65511	Erosional	Autumn	2	0	1.45	0.17	0.80	1.16	0.00	1.31
65511	Erosional	Autumn	1	0	1.47	0.00	2.11	4.03	0.00	3.33
65511	Depositional	Spring	1	0	13.85	0.00	8.62	4.82	0.00	3.79
65511	Depositional	Spring	2	2.5	0.00	0.06	0.00	8.88	2.26	0.02
65511	Erosional	Spring	1	10	4.09	0.00	0.30	6.57	0.00	3.14
65511	Erosional	Spring	2	5	1.64	1.42	0.00	1.05	0.00	0.00
67895	Depositional	Autumn	2	100	375.99	22.23	342.95	420.50	39.28	370.40

				Visual	Total	Organic	Inorganic	Total	Organic	Inorganic
		_		fines	surface (g	surface (g	surface (g	subsurface	subsurface	subsurface
Site	Patch	Season	Patch	(%)	m <sup>-2</sup> )	m⁻²)	_m⁻²)	(g m⁻²)	(g m⁻²)	(g m⁻²)
67895	Depositional	Autumn	1	85	103.47	5.57	91.88	191.64	18.61	167.01
67895	Erosional	Autumn	2	40	86.35	3.58	76.74	197.55	14.80	176.72
67895	Erosional	Autumn	1	5	71.48	6.11	60.88	99.84	7.43	87.92
67895	Depositional	Spring	1	100	544.25	54.68	470.58	1523.11	135.74	1368.39
67895	Depositional	Spring	2	100	709.02	81.75	603.82	2164.57	235.55	1905.57
67895	Erosional	Spring	1	30	36.90	0.00	31.50	292.31	33.23	251.64
67895	Erosional	Spring	2	30	73.33	0.00	65.54	251.16	11.80	230.25
81003	Depositional	Autumn	2	95	433.34	65.37	378.93	14.76	15.71	10.00
81003	Depositional	Autumn	1	95	101.46	27.39	83.02	1125.24	188.95	945.24
81003	Erosional	Autumn	2	85	278.26	69.17	217.43	1006.35	201.61	813.10
81003	Erosional	Autumn	1	80	39.22	19.55	27.85	534.44	101.17	441.44
81003	Depositional	Spring	1	100	1415.35	261.67	1146.95	9642.35	1716.75	7918.86
81003	Depositional	Spring	2	95	2369.20	488.35	1866.26	2521.77	495.76	2011.42
81003	Erosional	Spring	1	40	69.55	6.54	58.71	467.58	72.19	391.08
81003	Erosional	Spring	2	80	422.08	65.06	341.94	1056.47	162.60	878.80
155066	Depositional	Autumn	2	100	1068.56	276.74	791.83	1993.23	368.98	1624.25
155066	Depositional	Autumn	1	90	73.08	15.09	57.99	165.49	22.80	142.70
155066	Erosional	Autumn	2	75	863.79	248.20	615.59	1001.61	276.12	725.49
155066	Erosional	Autumn	1	85	50.28	11.00	39.28	158.31	28.70	129.60
155066	Depositional	Spring	1	80	168.13	24.20	135.25	406.76	51.53	346.55
155066	Depositional	Spring	2	100	258.03	31.41	213.44	622.62	21.10	588.35
155066	Erosional	Spring	1	80	386.49	59.89	319.55	628.00	84.94	536.01
155066	Erosional	Spring	2	70	595.06	135.01	453.08	616.48	139.81	469.70
161030	Depositional	Autumn	2	100	1978.37	544.03	1438.28	3130.35	843.12	2291.17
161030	Depositional	Autumn	1	97	330.53	38.19	300.09	326.19	52.56	281.39

				Visual	Total	Organic	Inorganic	Total	Organic	Inorganic
0.1	Detak	•	Detal	fines	surface (g	surface (g	surface (g	subsurface	subsurface	subsurface
Site	Patch	Season	Patch	(%)	m <sup>-</sup> 2)	m <sup>-</sup> 2)	m <sup>-</sup> 2)	(g m²)	(g m²)	(g m²)
161030	Erosional	Autumn	2	10	7.55	6.02	5.73	46.11	10.18	40.13
161030	Erosional	Autumn	1	5	8.61	2.42	8.76	91.50	16.49	77.58
161030	Depositional	Spring	1	100	2094.56	280.54	1804.96	6720.02	881.92	5829.04
161030	Depositional	Spring	2	80	542.05	32.05	476.15	756.53	46.95	675.73
161030	Erosional	Spring	1	50	26.21	0.00	21.92	162.69	14.19	138.93
161030	Erosional	Spring	2	33.3	99.71	0.05	87.88	292.57	48.48	232.30
161225	Depositional	Autumn	2	10	75.60	15.33	63.85	741.55	76.93	668.20
161225	Depositional	Autumn	1	10	172.19	18.46	156.95	813.64	64.82	752.04
161225	Erosional	Autumn	2	0	20.20	6.47	16.18	80.35	10.76	72.04
161225	Erosional	Autumn	1	60	351.26	15.22	338.89	631.90	26.93	607.83
161225	Depositional	Spring	1	90	248.89	7.84	226.37	721.15	53.65	652.82
161225	Depositional	Spring	2	50	64.71	1.93	57.47	381.65	25.39	350.96
161225	Erosional	Spring	1	10	174.62	6.27	154.76	469.20	18.69	436.93
161225	Erosional	Spring	2	5	38.81	0.00	38.61	114.20	3.37	100.07
162069	Depositional	Autumn	2	15	70.63	13.92	60.94	27.07	8.67	22.62
162069	Depositional	Autumn	1	100	783.50	181.93	605.08	1399.22	312.41	1090.32
162069	Erosional	Autumn	2	0	1.72	1.28	1.28	12.80	3.52	10.12
162069	Erosional	Autumn	1	0	5.41	2.07	4.97	21.37	5.06	17.94
162069	Depositional	Spring	1	0	7.52	0.00	8.68	40.40	1.32	38.15
162069	Depositional	Spring	2	10	103.92	10.57	90.87	157.42	29.80	125.15
162069	Erosional	Spring	1	0	6.99	0.00	8.02	12.81	0.00	12.78
162069	Erosional	Spring	2	0	7.65	0.00	7.60	15.58	0.00	16.66



#### Appendix 3.13 Diagnostic plots (sediment metrics models)





Figure A3.11 – residuals vs fitted (a), normal Q-Q (b), scale-location (c) and residuals vs leverage plot (d) for total sediment model.



Figure A3.12 – residuals vs fitted (a), normal Q-Q (b), scale-location (c) and residuals vs leverage plot (d) for visual fines linear model



Figure A3.13 – residuals vs fitted (a), normal Q-Q (b), scale-location (c) and residuals vs leverage plot (d) for background SSC model



Figure A3.14 – residuals vs fitted (a), normal Q-Q (b), scale-location (c) and residuals vs leverage plot (d) for organic surface model



Figure A3.15 – residuals vs fitted (a), normal Q-Q (b), scale-location (c) and residuals vs leverage plot (d) for inorganic surface model



Figure A3.16 – residuals vs fitted (a), normal Q-Q (b), scale-location (c) and residuals vs leverage plot (d) for total organic model



Figure A3.17 – residuals vs fitted (a), normal Q-Q (b), scale-location (c) and residuals vs leverage plot (d) for total inorganic model

# Appendix 3.14 Linear model results determining significant predictors of sediment metrics.

Response	Coefficient	Estimate	Std.	t	р
variable			Error		
	(Intercept)	9.607	1.648	5.828	<0.001*
	Bedrock	-0.025	0.012	-2.119	0.042*
Organic	Filamentous algae	0.292	0.143	2.041	0.050*
surface	Slope	-0.136	0.093	-1.457	0.155
	Background SSC	-0.025	0.006	-3.926	<0.001*
<b>df</b> 30	Coarse bed matrix	-0.010	0.005	-2.033	0.051
<b>Adj R<sup>2</sup></b> 0.810	Erosional flow	-0.020	0.004	-5.369	<0.001*
<b>F</b> 16.93	Q1	-0.620	0.206	-3.014	0.005*
<b>P</b> <0.001*	Q1090DF	1.392	0.568	2.451	0.020*
	Q50preWin	-1.631	0.497	-3.285	0.003*
	Stream power	0.690	0.184	3.759	0.001*
	Season (spring)	-0.904	0.231	-3.916	<0.001*
	(Intercept)	7.956	1.417	5.614	<0.001*
Inorganic	Width	-0.108	0.049	-2.211	0.034*
surface	Depth	0.029	0.018	1.605	0.118
	Bedrock	-0.042	0.017	-2.495	0.018*
<b>df</b> 32	Macrophyte	0.442	0.321	1.375	0.179
<b>Adj R<sup>2</sup></b> 0.726	Slope	0.333	0.134	2.485	0.018*
F 13.05	Coarse bed matrix	-0.026	0.008	-3.024	0.005*
<b>P</b> <0.001*	Erosional flow	-0.012	0.005	-2.367	0.024*
	Q1090DF	1.352	0.673	2.008	0.053
	Q50preSum	3.233	1.287	2.513	0.017*
	(Intercept)	13.980	2.362	5.919	<0.001*
	Depth	-0.049	0.015	-3.226	0.003*
	Bedrock	-0.062	0.016	-3.819	0.001*
Total organic	Filamentous algae	0.633	0.180	3.522	0.001*
	Coarse bed matrix	-0.028	0.007	-4.241	<0.001*
<b>df</b> 30	Erosional flow	-0.024	0.005	-5.020	<0.001*
<b>Adj R<sup>2</sup></b> 0.769	Q1	-0.835	0.265	-3.144	0.004*
<b>F</b> 13.41	Q1090DF	1.912	0.751	2.547	0.016*
<b>P</b> <0.001*	Q50preWin	-1.494	0.667	-2.239	0.033*
	Q20pre7d	1.492	0.902	1.654	0.108
	Stream power	0.480	0.246	1.952	0.060
	Season (spring)	-0.856	0.293	-2.919	0.007*

Table A3.9 – linear model results for other fine sediment metric responses. Significant coefficients are marked with an asterisk.

Response	Coefficient	Estimate	Std.	t	р
variable			Error		
	(Intercept)	10.098	1.208	8.358	<0.001*
Total	Width	-0.139	0.044	-3.152	0.004*
lotal	Bedrock	-0.065	0.016	-4.137	<0.001*
inorganic	Macrophyte	0.617	0.239	2.587	0.014*
df 32	Background	0.012	0.008	1.469	0.152
$\Delta di R^2 0.732$	Coarse bed matrix	-0.026	0.007	-3.561	0.001*
<b>F</b> 13 47	Q1090DF	1.420	0.653	2.175	0.037*
<b>P</b> <0.001*	Q20pre6m	0.870	0.655	1.329	0.193
	Q50preSum	3.500	1.285	2.724	0.010*
	Stream power	0.417	0.163	2.550	0.016*

### Appendix 3.15 EQR correlation matrices

	PSI EQR	EQR	DR											
PSI EQR	1	EPSIE	nixed EQ											
EPSI EQR	0.76*	1	EPSIn	QR										
EPSImixed EQR	0.77*	0.92*	1	oFSIE	EQR									
oFSI EQR	0.42	0.43	0.56*	1	ToFSI	EQR	ach							
ToFSI EQR	0.60*	0.49	0.67*	0.59*	1	CoFSI	fines re							
CoFSI EQR	-0.01	0.11	0.11	0.66*	-0.14	1	Visual	urface	Q					
Visual fines reach	-0.44	-0.41	-0.43	-0.20	-0.10	-0.14	1	Total s	c surfac	e				
Total surface	-0.37	-0.35	-0.38	-0.17	-0.07	-0.09	0.82*	1	Organi	nic surfa				
Organic surface	-0.32	-0.26	-0.30	0.01	-0.16	0.23	0.53*	0.65*	٦	Inorga	ediment			
Inorganic surface	-0.34	-0.33	-0.36	-0.15	-0.03	-0.10	0.82*	0.99*	0.63*	1	Total s	rganic	101	
Total sediment	-0.20	-0.21	-0.21	0.01	0.01	0.04	0.73*	0.92*	0.56*	0.91*	1	Total o	norganic	Ŋ
Total organic	-0.28	-0.28	-0.28	0.04	-0.16	0.26	0.62*	0.74*	0.89*	0.73*	0.76*	1	Total ir	S puno.
Total inorganic	-0.18	-0.19	-0.20	0.01	0.04	0.02	0.73*	0.92*	0.55*	0.92*	1.00*	0.75*	1	Backgi
Background SSC	-0.19	-0.12	-0.15	-0.11	-0.16	-0.05	0.22	0.23	-0.21	0.23	0.36	0.06	0.34	1

Figure A3.18 – Correlation matrix of EQRs of sediment-specific biomonitoring index scores with different metrics of fine sediment.

	WHPT ASPT EQR	. Ntaxa EQR																			
WHPT ASPT EQR	1	WHPT	P EQR																		
WHPT Ntaxa EQR	0.49	1	BMW	EQR																	
BMWP EQR	0.68*	0.93*	1	Ntaxa	- EQR																
Ntaxa EQR	0.48	0.97*	0.94*	1	ASPT	EQR															
ASPT EQR	0.90*	0.52	0.75*	0.52	1	LIFE	EQR	EQR													
LIFE EQR	0.40	0.08	0.27	0.19	0.31	1	EPT	dance	QR												
EPT EQR	0.53	0.39	0.54*	0.44	0.62*	0.25	1	Abun	non			A									
Abundance EQR	-0.03	0.29	0.25	0.36	-0.08	0.47	-0.01	1	Shan	EQR		age EC									
Shannon EQR	0.21	0.49	0.48	0.48	0.28	-0.13	0.40	-0.36	1	FRic	EQR	ercenta	R								
FRic EQR	0.36	0.91*	0.76*	0.85*	0.36	-0.01	0.19	0.31	0.42	1	FDis	dder p	er EQF								
FDis EQR	-0.05	0.12	0.13	0.09	0.02	-0.20	0.09	-0.49	0.54	0.10	1	Shre	hredde	tch							
Shredder percentage EQR	0.58*	0.31	0.45	0.35	0.49	0.45	0.14	0.24	0.11	0.26	-0.30	1	CWMs	fines rea							
CWM shredder EQR	0.50	0.30	0.41	0.34	0.44	0.36	0.09	0.31	0.02	0.28	-0.43	0.95*	1	Visual	surface	face					
Visual fines reach	0.00	-0.12	-0.11	-0.17	0.06	-0.34	-0.29	-0.51	0.07	-0.07	0.10	-0.02	-0.03	1	Total	nic sur	urface				
Total surface	-0.01	-0.35	-0.30	-0.39	0.02	-0.29	-0.25	-0.56*	-0.10	-0.32	-0.09	-0.06	-0.06	0.82*	1	Orga	anic sı	ent			
Organic surface	0.04	-0.26	-0.29	-0.36	-0.03	-0.29	-0.27	-0.40	-0.11	-0.14	0.10	-0.20	-0.24	0.53	0.65*	1	Inorg	sedim	0		
Inorganic surface	0.02	-0.35	-0.29	-0.39	0.05	-0.27	-0.23	-0.57*	-0.07	-0.33	-0.06	-0.04	-0.05	0.82*	0.99*	0.63*	1	Total	organ	nic	SC
Total sediment	0.02	-0.33	-0.29	-0.35	-0.01	-0.12	-0.26	-0.41	-0.09	-0.27	-0.26	0.11	0.12	0.73*	0.92*	0.56*	0.91*	1	Total	inorga	S pund S
Total organic	-0.03	-0.28	-0.33	-0.37	-0.12	-0.24	-0.38	-0.36	-0.08	-0.14	-0.07	-0.07	-0.09	0.62*	0.74*	0.89*	0.73*	0.76*	1	Total	lackgro
Total inorganic	0.05	-0.33	-0.29	-0.36	0.01	-0.10	-0.24	-0.43	-0.08	-0.28	-0.25	0.11	0.11	0.73*	0.92*	0.55*	0.92*	1.00*	0.75*	1	ш
Background SSC	-0.27	-0.09	-0.11	-0.02	-0.20	0.13	-0.26	0.04	0.02	-0.07	-0.17	0.02	0.06	0.22	0.23	-0.21	0.23	0.36	0.06	0.34	1

Figure A3.19 – Correlation matrix of EQRs of other biomonitoring indices with different metrics of fine sediment.

# Appendix 3.16 Linear model results determining significant predictors of biomonitoring indices.

Table A3.10 - Linear model results for each fine sediment metric. Significant effects are indicated with an asterisk.

Response	Coefficient	Estimate	Std.	Т	р
variable			Error	value	
	(Intercept)	4.362	0.341	12.808	<0.001*
- 501	Q1090DF	-0.415	0.171	-2.424	0.021*
0F51	Q20pre6m	-0.631	0.212	-2.983	0.005*
46.20	Q50preSum	-1.231	0.418	-2.944	0.006*
	Q20pre7d	-0.480	0.303	-1.584	0.123
<b>AUJ</b> $\mathbf{R}^{-}$ 0.031	Detritus	-0.144	0.045	-3.179	0.003*
P = 0.790	Filamentous algae	-0.075	0.046	-1.631	0.113
<b>P</b> < 0.001	Slope	0.208	0.050	4.172	<0.001*
	Stream power	-0.238	0.059	-4.049	<0.001*
	Season (spring)	0.295	0.089	3.319	0.002*
	(Intercept)	5.810	0.388	14.985	<0.001*
	Q1090DF	0.600	0.200	2.996	0.005*
	Q20pre6m	0.327	0.182	1.791	0.083
ToFSI	Q50preWin	-0.353	0.169	-2.088	0.045*
	Q50preSum	0.707	0.338	2.093	0.045*
<b>df</b> 30	End Group	-0.176	0.064	-2.766	0.010*
<b>Adj R</b> <sup>2</sup> 0.480	Depth	-0.225	0.049	-4.567	<0.001*
<b>F</b> 4.442	Bedrock	-0.151	0.041	-3.637	0.001*
<b>p</b> 0.001*	Altitude	-0.082	0.046	-1.807	0.081
	Slope	-0.065	0.051	-1.268	0.215
	Distance from source	0.111	0.068	1.644	0.111
	Stream power	0.197	0.067	2.948	0.006*
WHPT_ASPT	(Intercept)	2.910	0.706	4.122	<0.001*
	Total inorganic	-0.193	0.102	-1.900	0.068
<b>df</b> 31	Background	-0.132	0.101	-1.313	0.200
<b>Adj R</b> <sup>2</sup> 0.577	Q1090DF	-1.147	0.374	-3.071	0.005*
<b>F</b> 4.999	Q20pre6m	-0.973	0.386	-2.521	0.018*

Response	Coefficient	Estimate	Std.	Т	р
variable			Error	value	
<b>p</b> <0.001*	Q50preWin	0.816	0.378	2.158	0.040*
	Q20pre7d	-1.106	0.487	-2.272	0.031*
	ASL	0.240	0.089	2.684	0.012*
	Depth	0.191	0.108	1.771	0.088
	Detritus	-0.214	0.087	-2.458	0.021*
	Filamentous algae	-0.164	0.085	-1.939	0.063
	Slope	0.320	0.097	3.301	0.003*
	Distance from source	-0.213	0.105	-2.026	0.053
	Stream power	-0.508	0.145	-3.511	0.002*
	Season (spring)	0.363	0.169	2.142	0.041*
	(Intercept)	5.238	5.624	0.931	0.359
	Total inorganic	-2.463	0.761	-3.236	0.003*
WHPT_NTAXA	Background SSC	1.417	0.805	1.759	0.088
	Q1090DF	-6.002	2.626	-2.285	0.029*
<b>df</b> 31	Q20pre6m	7.211	3.280	2.198	0.036*
<b>Adj R</b> <sup>2</sup> 0.524	Q50preSum	-14.503	7.366	-1.969	0.058
<b>F</b> 5.517	Asl	1.149	0.913	1.258	0.218
<b>p</b> <0.001*	Filamentous algae	-0.919	0.681	-1.350	0.187
	Altitude	-1.342	0.750	-1.788	0.083
	Slope	-1.281	0.767	-1.670	0.105
	Stream power	-1.456	0.792	-1.838	0.076
	(Intercept)	6.837	0.172	39.764	<0.001*
	Total inorganic	-0.150	0.058	-2.575	0.015*
	Q20pre6m	-0.735	0.267	-2.755	0.010*
<b>df</b> 32	Q50preWin	-0.331	0.163	-2.036	0.050*
<b>Adi</b> $\mathbf{R}^2 \cap 550$	Q20pre7d	-0.865	0.306	-2.830	0.008*
<b>F</b> 6 547	Bedrock	-0.113	0.057	-1.998	0.054
<b>p</b> < 0.001*	D1etritus	-0.114	0.059	-1.946	0.060
	Altitude	0.165	0.061	2.723	0.010*
	Slope	0.188	0.060	3.127	0.004*
	Season (spring)	0.298	0.112	2.665	0.012*

Response	Coefficient	Estimate	Std.	Т	р
variable			Error	value	
	(Intercept)	-21.922	13.527	-1.621	0.118
	Background SSC	-2.636	1.766	-1.493	0.148
	Q1090DF	-24.809	7.044	-3.522	0.002*
	Q20pre6m	-12.815	7.684	-1.668	0.108
	Q50preWin	14.092	6.466	2.179	0.039*
EPT	Q20pre7d	-24.884	9.139	-2.723	0.012*
	Asl	3.594	1.620	2.218	0.036*
<b>df</b> 25	Depth	4.598	1.913	2.403	0.024*
<b>Adj R²</b> 0.579	Bedrock	1.575	1.401	1.124	0.272
<b>F</b> 4.519	Detritus	-2.226	1.522	-1.462	0.156
<b>p</b> <0.001*	Filamentous algae	-4.475	1.600	-2.797	0.010*
	Altitude	3.177	1.703	1.866	0.074
	Slope	2.627	1.895	1.386	0.178
	Distance from source	-4.215	2.075	-2.031	0.053
	Coarse bed matrix	4.532	2.045	2.216	0.036*
	Stream power	-6.729	2.568	-2.620	0.015*
	Season (spring)	7.115	3.049	2.333	0.028*
	(Intercept)	-0.014	0.003	-4.232	<0.001*
EDio	Total inorganic	-0.004	0.001	-5.229	<0.001*
	Background SSC	0.001	0.001	1.472	0.151
<b>df</b> 33	Q1090DF	-0.011	0.002	-5.363	<0.001*
<b>Adi R</b> <sup>2</sup> 0 669	Q20pre6m	0.006	0.002	2.689	0.011*
<b>F</b> 11 35	depth	0.003	0.001	4.891	<0.001*
<b>n</b> <0.001*	Slope	-0.002	0.001	-3.326	0.002*
	Coarse bed matrix	-0.002	0.001	-2.538	0.016*
	Season (spring)	-0.002	0.001	-1.620	0.115
FDis	(Intercept)	3.170	0.943	3.360	0.002*
	Organic surface	0.332	0.159	2.089	0.045*
<b>df</b> 31	Background SSC	0.219	0.173	1.267	0.214
<b>Adj R²</b> 0.450	Q1090DF	-1.344	0.544	-2.470	0.019*
<b>F</b> 4.36	Q50preWin	1.720	0.622	2.766	0.009*

Response	Coefficient	Estimate	Std.	Т	р
variable			Error	value	
<b>p</b> 0.001*	Depth	0.376	0.178	2.116	0.042*
	Bedrock	0.294	0.137	2.155	0.039*
	Detritus	-0.324	0.148	-2.181	0.037*
	Filamentous algae	-0.569	0.149	-3.804	0.001*
	Macrophyte	-0.276	0.155	-1.783	0.084
	Stream power	-0.396	0.215	-1.842	0.075

# Appendix 3.17 Diagnostic plots of linear models determining significant predictors of biomonitoring indices.



Figure A3.20 – residuals vs fitted (a), normal Q-Q (b), scale-location (c) and residuals vs leverage plot (d) for PSI model



Figure A3.21 – residuals vs fitted (a), normal Q-Q (b), scale-location (c) and residuals vs leverage plot (d) for EPSI model



Figure A3.22 – residuals vs fitted (a), normal Q-Q (b), scale-location (c) and residuals vs leverage plot (d) for EPSImixed model



Figure A3.23 – residuals vs fitted (a), normal Q-Q (b), scale-location (c) and residuals vs leverage plot (d) for CoFSI model



Figure A3.24 – residuals vs fitted (a), normal Q-Q (b), scale-location (c) and residuals vs leverage plot (d) for oFSI model



Figure A3.25 – residuals vs fitted (a), normal Q-Q (b), scale-location (c) and residuals vs leverage plot (d) for ToFSI model



Figure A3.26 – residuals vs fitted (a), normal Q-Q (b), scale-location (c) and residuals vs leverage plot (d) for WHPT ASPT model



Figure A3.27 – residuals vs fitted (a), normal Q-Q (b), scale-location (c) and residuals vs leverage plot (d) for WHPT\_NTAXA model



Figure A3.28 – residuals vs fitted (a), normal Q-Q (b), scale-location (c) and residuals vs leverage plot (d) for LIFE model



Figure A3.29 – residuals vs fitted (a), normal Q-Q (b), scale-location (c) and residuals vs leverage plot (d) for EPT model



Figure A3.30 – residuals vs fitted (a), normal Q-Q (b), scale-location (c) and residuals vs leverage plot (d) for FRic model



Figure A3.31 – residuals vs fitted (a), normal Q-Q (b), scale-location (c) and residuals vs leverage plot (d) for FDis model



Appendix 3.18 Scree plot for RLQ

Figure A3.32 – A scree plot showing the contribution of each dimension to the total explained variance for the RLQ analysis

### **Appendix 4 – Ethics documentation**

#### Appendix 4.1 Ethics certificate P40594



### **Certificate of Ethical Approval**

Applicant:

Morwenna McKenzie

Project Title:

Quantifying the response of macro invertebrates to gradients of fine sediment pollution

This is to certify that the above named applicant has completed the Coventry University Ethical Approval process and their project has been confirmed and approved as Low Risk

Date of approval:

11 January 2016

Project Reference Number:

P40594

#### Appendix 4.2 Ethics certificate P45642



### **Certificate of Ethical Approval**

Applicant:

Morwenna McKenzie

Project Title:

Quantifying the response of macroinvertebrates to gradients of fine sediment pollution

This is to certify that the above named applicant has completed the Coventry University Ethical Approval process and their project has been confirmed and approved as Medium Risk

Date of approval:

09 September 2016

Project Reference Number:

P45642

#### Appendix 4.3 Ethics certificate P52533



### **Certificate of Ethical Approval**

Applicant:

Morwenna McKenzie

Project Title:

Improving the knowledge base on the physical effects of fine sediment for macroinvertebrates

This is to certify that the above named applicant has completed the Coventry University Ethical Approval process and their project has been confirmed and approved as Medium Risk

Date of approval:

20 July 2017

Project Reference Number:

P52533