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QUANTIFYING SUBMERGED DEPOSITED FINE SEDIMENTS IN RIVERS AND STREAMS USING DIGITAL IMAGE ANALYSIS

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ABSTRACT

Deposited fine sediment is an essential component of freshwater ecosystems. Nonetheless, anthropogenic activities can modify natural fine sediment levels, impacting the physical, chemical and biological characteristics of these ecosystems. An ability to quantify deposited fine sediment is critical to understanding its impacts and successfully managing the anthropogenic activities that are responsible for modifying it. One widely used method, the visual estimate technique, relies on subjective estimates of particle size and percentage cover. In this paper, we present two novel alternative approaches, based on non-automated digital image analysis (DIA), which are designed to reduce the subjectivity of submerged and surficial fine sediment estimates, and provide a verifiable record of the conditions at the time of sampling. The DIA methods were tested across five systematically selected, contrasting temperate stream and river typologies, over three seasons of monitoring. The resultant sediment metrics were strongly, positively correlated with visual estimates ($r_s = 0.90$, and $r_s = 0.82$, $p < 0.01$), and similarly strongly, but negatively correlated with a sediment-specific biotic index, suggesting some degree of biological relevance. The DIA technique has the potential to be a valuable tool for application in numerous areas of river research, where a non-destructive, less subjective and verifiable method is desirable. Copyright © 2016 The Authors River Research and Applications Published by John Wiley & Sons Ltd

KEY WORDS: deposited fine sediment; visual estimates; habitat assessment; digital image analysis; river substrate

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INTRODUCTION

Fine sediment (organic and inorganic particles; < 2 mm) is an essential component of freshwater ecosystems (Owens *et al.*, 2001). Nonetheless, anthropogenic activities can lead to modified levels of fine sediment delivery to surface waters, impacting the physical, chemical and biological characteristics of these ecosystems (Wood and Armitage, 1997). An ability to quantify deposited fine sediment is critical to understanding its impacts and successfully managing the anthropogenic activities that are responsible for modifying it. A number of methods for quantifying both surficial (Table I) and subsurface fine sediment have been documented. Some of the most commonly used methods by both researchers and monitoring agencies are rapid assessments (Faustini and Kaufmann, 2007), often comprising visual estimates of substrate composition (Descloux *et al.*, 2010; Sennatt *et al.*, 2006). These assessments involve operators estimating the surficial fine sediment in terms of the percentage cover of different particle sizes across a site. Frequently, the particle size classifications follow the Wentworth system (Wentworth,

1922): boulders (≥ 256 mm), cobbles (64–256 mm), gravels/pebbles (2–64 mm), sand (< 2 mm ≥ 0.06 mm), silt and clay (< 0.06 mm) (Bain and Stevenson, 1999; Clapcott *et al.*, 2011; Environment Agency, 2003). Surface percentage cover estimates often rely on the subjective ability of the operator to identify particle sizes and percentage cover of these particles (Faustini and Kaufmann, 2007). Whilst not necessarily an indicator of subsurface fine sediment, percentage cover is likely to be important in terms of determining the biological implications of altered fine sediment dynamics, as it is concerned with the surface ‘drape’, which can lower the oxygen availability in the benthos and reduce the quantity of forage and refugial habitat (Sutherland *et al.*, 2010).

Research has investigated the quality of these methods (Clapcott *et al.*, 2011; Roper *et al.*, 2002; Sennatt *et al.*, 2006; Whitacre *et al.*, 2007). In terms of precision (i.e. the degree of closeness between repeated measures), visual estimates by 10 operators have been shown to vary by up to 40 percentage points for sites with the same Wolman pebble count (Clapcott *et al.*, 2011), although this is likely to be an extreme example as operators received no prior training on the technique. Furthermore, the Wolman pebble count has been shown as biased against small particles (Diplas and Lohani, 1997; Marcus *et al.*, 1995). In terms of accuracy (i.e. closeness to the true value), the ‘true’ value for

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Table I. Various approaches used globally to quantify submerged, surficial fine sediments in rivers and streams

Approach	Description	Reference
Particle counts	<ul style="list-style-type: none"> • Wolman pebble count. Particles are selected at random, by sampling using toe-to-heel spacing and selecting the first particle touched by a finger at the toe of the operator's boot. Transects between bankfull widths within habitats of interest (or a zigzag pattern) are followed until 100 particles (or desired number) are selected. 	Wolman, 1954; Bevenger and King, 1995; Diplas and Lohani, 1997
	<ul style="list-style-type: none"> • Wolman pebble count (modified). Grid-by-number methods using a measuring tape, wire mesh or frame with elastic bands to select particles. 	Kellerhals and Bray, 1971; Bunte and Abt, 2001
	<ul style="list-style-type: none"> • Wolman pebble count (modified). The toe-to-heel approach is used in conjunction with a clay disc and piston, which is pressed onto the streambed to sample fine sediment. 	Fripp and Diplas, 1993)
In-stream visual estimate	<ul style="list-style-type: none"> • A measuring tape is placed between the beginning and end of a transect. At 0.3 m increments, the dominant sediment class over the length is recorded. • Using an underwater viewer, the percentage cover of fine sediment is estimated. 	Platts <i>et al.</i> , 1983 Matthaei <i>et al.</i> , 2006
Bankside visual estimate	<ul style="list-style-type: none"> • Substrate composition is estimated following a visual inspection of the reach from the stream bank. 	Environment Agency, 2003
Photographic image analysis	<ul style="list-style-type: none"> • The percentage of fine sediment is estimated from the stream bank. 	Clapcott <i>et al.</i> , 2011
	<ul style="list-style-type: none"> • Above water image capture. Areas of substrate are photographed using a photographic film camera, an underwater viewer and a structure to isolate the area of streambed. Photographic transparencies were then projected at three times life size and onto a grid with 400 squares. The predominant particle size in every fourth square was then recorded. 	Gee, 1979
	<ul style="list-style-type: none"> • Similar to the technique used by Gee (1979), with the addition of a digitizing program to obtain the particle size distribution. 	Ibbeken and Schleyer, 1986
	<ul style="list-style-type: none"> • Areas of substrate are photographed from above the water using a photographic film camera and underwater viewer. Photographic transparencies are digitized and analysed using Geographic Information Software. 	Whitman <i>et al.</i> , 2003
Artificial mats	<ul style="list-style-type: none"> • Images are collected using a modified camera with underwater housing and a light ring. The resulting images are approximately 0.02 m across, and are analysed using an autocorrelation algorithm to determine grain sizes. 	Rubin <i>et al.</i> , 2007
	<ul style="list-style-type: none"> • Artificial turf mats (0.15 m × 0.10 m) are fixed to the streambed and left for 3 weeks. Mats are carefully retrieved and placed in zip-lock bags and returned to the laboratory where the sediment is washed out, sieved and collected for drying and weighing. 	Von Bertrab <i>et al.</i> , 2013
Adhesive plates	<ul style="list-style-type: none"> • A plate is covered with a thin layer of adhesive material (i.e. clay) and is pressed onto the streambed. The sample is then wet-sieved to remove the clay. 	Fripp and Diplas, 1993
Resuspension	<ul style="list-style-type: none"> • A steel cylinder measuring 1 m in height is pushed 0.1 m into the streambed. The water within the cylinder is agitated, artificially suspending the surficial fine sediment, allowing for three 0.5 L samples to be collected. This process is carried out at three or more 'representative' sites, and samples are used to determine the mean mass (g m^{-2}) of sediment released. 	Lambert and Walling, 1988
	<ul style="list-style-type: none"> • Modified from Lambert and Walling (1988), using a steel cylinder measuring 0.75 m in height. Samples of 0.05 L are collected from two 'depositional' and two 'erosional' patches at each site. 	Duerdoth <i>et al.</i> , 2015
	<ul style="list-style-type: none"> • Qualitative assessment using five categories relating to the percentage that large particles were covered by fine sediment. 	Platts <i>et al.</i> , 1983
Embeddedness	<ul style="list-style-type: none"> • Qualitative assessment of embeddedness using three categories: 1 = lying loosely on top of the bed, 2 = partly covered by surrounding substratum, 3 = well buried in the surrounding substratum or firmly wedged in by surrounding stones. 	Matthaei <i>et al.</i> , 1999
	<ul style="list-style-type: none"> • Assessment of embeddedness over 11 transects (55 particles) by estimating the percentage embeddedness of each particle. 	Peck <i>et al.</i> , 2002

substrate composition in a stream is generally unknown (Sutherland *et al.*, 2010), although some authors have used 'measured' methods in place of a true value. However, it

is important to remember that the 'measured' method is not necessarily closer to the 'true' value, and a difference between the estimated and 'measured' methods will always

occur because of the different methodologies measuring slightly different aspects of the substrate and any fine sediment.

An alternative to the conventional method of visual estimates of substrate composition is the use of image-based techniques. Photography has been used to some extent by geomorphologists and sedimentologists for over 35 years (Adams, 1979), with recent work utilizing technological advances to determine grain size characteristics, using (i) automated grain size analysis of images and (ii) geostatistical techniques and empirical calibration, mainly of exposed gravel bed rivers (e.g. Buscombe, 2008; Butler *et al.*, 2001; Carbonneau *et al.*, 2004; Graham *et al.*, 2005b; Graham *et al.*, 2005a; McEwan *et al.*, 2000; Rice, 1995; Sime and Ferguson, 2003; Warrick *et al.*, 2009). Both Rubin *et al.* (2007) and Buscombe *et al.* (2010) have applied their techniques underwater, the latter in controlled conditions. Whilst these techniques provide opportunities to process large numbers of images and obtain particle size distributions, the identification of fine sediments, particularly submerged, and/or cohesive sediments, is often limited because of technological limitations and image resolutions (Bertoldi *et al.*, 2012; Graham *et al.*, 2005a) as well as a requirement for well-defined grain boundaries (Graham *et al.*, 2005a; McEwan *et al.*, 2000).

In addition to the precision and accuracy of each technique, the biological relevance of the resulting sediment metric is often of great concern. In the European Union, whilst sediment deposition is not directly legislated, as part of the European Union Water Framework Directive member states are required to achieve 'Good Ecological Status' in surface waters. Given the ecological impacts of fine sediment (Bilotta and Brazier, 2008), a method of quantifying fine sediment that provides a biologically relevant metric is therefore highly desirable.

The aim of this paper is to provide 'proof of concept' for two novel, non-automated, digital image analysis (DIA) methods for quantifying submerged, surficial deposited fine sediment, building on the work from previous authors, many of whom were attempting to characterize complete grain size distributions. Crucially, the techniques have to be (i) applicable to fine sediments on submerged river/streambeds; (ii) more objective and verifiable than existing visual estimate methods; and (iii) simple to carry out requiring little specialist software or training. The results from these new procedures are compared with those based on visual estimates. The precision of each method is assessed using an independent operator, and the biological relevance is evaluated using the relationship between each sediment metric and the invertebrate community composition, quantified using a sediment-specific biotic index (Turley *et al.*, 2016). Between-transect and between-season variation is also assessed to provide an insight into fine sediment spatial

variation and the likely ability of the DIA sampling design to characterize fine sediment conditions across the site.

METHODS

Site selection

Five sites were selected from a database of 835 reference condition or minimally impacted sites on rivers and streams located throughout the UK that were sampled between 1978 and 2004 [RIVPACS IV—River Invertebrate Prediction and Classification System—NERC (CEH) 2006; May 2011 version. Database rights NERC (CEH) 2006 all rights reserved]. The five sites represented a range of different river types, with contrasting environmental characteristics; depths between 0.05 m and 0.80 m and importantly, a range of fine sediment covers (0–55% based on visual estimates). The site selection process involved both stratified and systematic stages to ensure sites with a range of deposited fine sediment conditions were selected with minimal bias. Briefly, this involved ranking the 835 sites in terms of the percentage of the substrate comprised fine sediment (based on previous visual estimates) and selecting those sites that represented the minimum, 25th, 50th, 75th and 100th percentile values. Using satellite imagery, a number of criteria were then applied to the selected sites to minimize selection bias, to ensure that sites were relatively unimpacted by point source organic pollution (e.g. no sewage treatment works and significant urban/rural dwellings upstream), physical modifications (e.g. no obvious channelization) or arable agriculture (and the associated fertilizers and pesticides) and to reduce the number of pressures with the potential to influence the macroinvertebrate community composition. If a site failed to meet the criteria it was rejected and the next ranked site was put through the same criteria, until five sites were found that met all criteria. The characteristics of these systematically selected sites are shown in Table II. In this study,

Table II. Characteristics of the five stream sites, based on data from the RIVPACS IV database

Environmental variables	Site 1	Site 2	Site 3	Site 4	Site 5
Altitude ^a (m)	364	205	75	25	66
Slope ^a (m m ⁻¹)	0.01	0.008	0.008	0.004	0.003
Average width ^b (m)	1.9	5.8	3.9	8.0	11.8
Average depth ^b (m)	0.08	0.30	0.14	0.25	0.65
Alkalinity ^c	24.4	6.8	61.8	111.7	247.0
Fine sediment ^b (%)	0	12	18	34	55

Environmental variables derived from the following:

^amap data,

^bmean of three seasonal measurements,

^cmean of 12 monthly measurements.

each site consisted of a short river reach, approximately 10 m in length, over which the visual estimates, digital images and macroinvertebrates were collected.

Visual estimates

The percentage of fine sediment cover was estimated over the site and corresponded with the area that was sampled for invertebrates, following the UK Environment Agency protocol (Environment Agency, 2003). Briefly, this involved observations carried out from the stream bank, and where necessary, probing areas of substrate. The overall percentage of fine sediment (<2 mm) was recorded to the nearest 5%.

Digital image collection

In order to reduce the potential for sampling bias (i.e. selecting areas of high or low sediment), each site was photographed systematically using equidistant transects. Five transects were set up at each site, beginning at the downstream end of the site and positioned every 2 m (see Supporting Information), incorporating riffles, runs and glides where present. Three sampling locations were positioned on each transect at $\frac{1}{4}$, $\frac{1}{2}$ and $\frac{3}{4}$ of the wetted channel width. Each site was sampled over three seasons (spring, summer and autumn) in the same year (2014).

A relatively low cost (<500 GBP) 16.1-megapixel waterproof digital camera (*Panasonic DMC-FT5*) was used to eliminate the need for a structure to protect the camera from water. In addition to being waterproof, another important feature was the camera's reported performance under low light conditions and its minimum focal distance. Both of these were important considerations as photographs were to be taken within the water column, which would result in reduced natural light and a limited distance between the camera and the streambed.

At each site and on each visit, the camera was submerged and test photographs were taken and reviewed *in situ* to determine the approximate, optimum distance between the camera and the streambed. The camera was held from a vertical position to capture images of the substrate in plan view. The optimum distance was deemed as that which resulted in an image that captured the maximum area of the streambed (which was limited by the water depth) whilst providing a suitably clear image for later analysis. In streams that are relatively turbid, this can result in a small area (<0.01 m²) of streambed being captured in each image. Beginning at the downstream end of the site, so as not to disturb the substrate prior to image capture, an image was taken at each sampling location, with image quality being briefly reviewed before moving on to the next location. The distance between the streambed and the camera lens was measured using a

graduated measuring pole with 0.01 m increments and was recorded for each image. The minimum and maximum distances between the camera lens and the streambed in this study were 0.02 and 0.76 m, respectively. Once the images had been collected, they were returned to the laboratory and uploaded for processing.

Image processing

The image processing was kept to a minimum in comparison with the automated processes described by previous authors, and was carried out using Adobe Photoshop Elements 13, but can also be conducted using image editing freeware (e.g. Pixlr 3.0). The relationship between camera height and the area (m²) of streambed captured in the image was determined using a tripod setup in the laboratory. Images of a 1 m rule were captured with the camera positioned at 0.05 m height increments, between 0.02 and 0.80 m, and the length of the rule captured in each image, at each height was recorded. Using these data, a formula was derived using linear regression to calculate the length (L ; metres) of streambed captured for each camera height (h ; metres) increment between 0.02 and 0.80 m using the *Panasonic DMC-FT5*, Panasonic Corporation, Osaka, Japan. assuming a planar bed (Equation 1). The length of each image was then resized so that they represented the actual captured area of the streambed, and their resolution was set to 180 pixels per inch.

$$L = 1.223h + 0.018 \quad (1)$$

Estimate-based digital image analysis

Images collected for DIA were first used by the operator to estimate the percentage of fine sediment. This involved a grid layout (10×10) being placed over the image in Photoshop (each square of the grid representing 1% of the area in the image) to aid a systematic summation. In order to assist in the identification of fine sediment, the *Brush Tool* was set to 12 pixels as this size brush represents approximately 2 mm at the resolution of the saved images, allowing a comparison between the cursor and particles. Where necessary, the contrast and brightness of the image was manipulated to improve the clarity of dark areas of substrate (e.g. interstices). The percentage of fine sediment was then estimated to the nearest 1% and recorded. This process required less than 5 min per image.

Software-based digital image analysis

Using the images collected in the field, areas of fine sediment were highlighted and quantified. This involved firstly opening the resized images in Photoshop and selecting the foreground colour of 'ff0000' along with the *Brush Tool* set at 14 pixels. Moving the *Brush Tool*

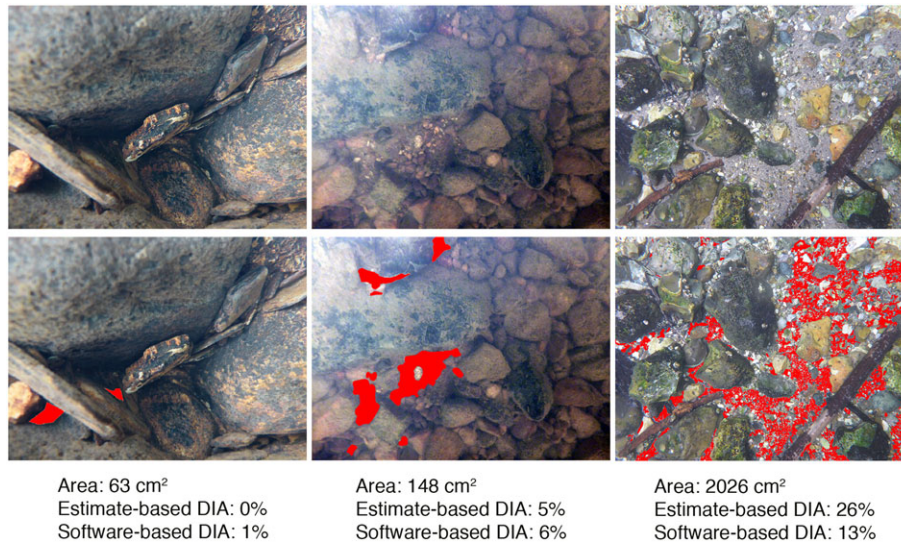


Figure 1. Examples of digital image analysis (DIA) images (top row) and these images with fine sediment highlighted (bottom row). Area of streambed and fine sediment estimates based on estimate-based DIA and software-based DIA are detailed. This figure is available in colour online at wileyonlinelibrary.com/journal/rra

over the image, any particles that were less than the size of the cursor were then highlighted with the foreground colour (Figure 1) using either the *Brush Tool*, *Polygonal Lasso* or in some cases the *Magic Wand Tool*. The *Magic Wand Tool* could only be used at a low tolerance (<25%) when the areas of fine sediment were suitably contrasted to the other particles surrounding it. Once the fine sediment had been identified and highlighted (requiring up to 30 min per image), the images were ‘saved for web’, as Portable Network Graphics. The images were then uploaded to *PixelCount*, a freeware Google Chrome application that was developed to count the number of pixels of a specific colour (ff0000—which is set prior to uploading the images). The application outputs the proportion of each image that matches the chosen colour. By converting this proportion to an area (m²) using the known area of the streambed in each image, the total area (m²) of fine sediment in all 15 images was divided by the total overall area in the images and multiplied by 100 to obtain the overall percentage cover of fine sediment (Equation 2).

$$\% \text{ fine sediment} = \frac{\sum(a \cdot p)}{\sum a} * 100 \quad (2)$$

In Equation 2, a is the total area (m²) of streambed in the image and p is the proportion of the image that is highlighted. In the numerator of the equation, the area in each image is multiplied by the proportion of highlighted fine sediment in the image, and the products are then summed. In the denominator, the areas of each image

are summed. The equation takes into consideration that each image can capture a different size area, and as such, the same proportion of fine sediment in any two images may not represent the same size area.

Precision of digital image analysis techniques

In order to evaluate the precision of the DIA techniques, an independent operator carried out duplicate analyses for a single season for each of the five sites. The exact seasonal sample that was analysed was randomly selected, and inter-operator comparisons were made between the same sites and seasons. Whilst the first operator had knowledge of the sites (having visited the location and conducted visual estimates), the independent operator had no prior knowledge of the site characteristics. Brief training (1 h) was provided using 10 trial images with a range of sediment covers, to guide the independent operator through the process of identifying, estimating and highlighting fine sediment in the images.

Fine sediment spatial variation

To evaluate the effectiveness of the image collection sampling design as well as the spatial variation in fine sediment at the study sites, the between-transect variation in fine sediment values were assessed in terms of the 95% confidence intervals for the mean percentage of fine sediment based on five transects. This required the percentage of fine sediment cover in each one of the sites five transects to be calculated individually (using the three images in each transect) and averaged over the five transects.

Biological relevance

In this study, we assess the biological relevance of the methods by comparing their Spearman's rank correlation coefficients with a sediment-specific biotic index; the mixed taxonomic level, Empirically-weighted Proportion of Sediment-sensitive Invertebrates (E-PSI) index (Turley *et al.*, 2016). The E-PSI index is designed to identify the impacts of fine sediment pressures in streams using the benthic invertebrate community composition. Invertebrates were initially rated by their sensitivity to fine sediment, based on expert knowledge and an assessment of biological and ecological traits that result in a sensitivity or tolerance of fine sediment (Extence *et al.*, 2011; Turley *et al.*, 2014). The E-PSI index maintains this biological basis and assigns detailed sensitivity weights based on empirical data of invertebrate community compositions and visual estimates of fine sediment (Turley *et al.*, 2016). For this reason, the index may be more strongly correlated with visual estimates than with the other methods. The invertebrate data used to calculate this index were collected using the UK standard method: a standardized 3-min kick sample using a 900- μm mesh hand net, followed by a 1-min hand search. All in-stream habitats identified at the site were sampled in proportion to their occurrence (Environment Agency, 2009). Invertebrates were recorded to the lowest practicable taxonomic level, mostly to species or genus, with the exception of Oligochaeta (class) and Diptera (family).

Statistical analysis

The data were compiled in Microsoft Excel and analysed using SPSS statistical software (IBM SPSS Statistics 22). Fine sediment data were aligned with E-PSI scores that were calculated using mixed taxonomic level data collected contemporaneously. The fine sediment data did not satisfy the assumption of bivariate normality for Pearson's correlation coefficient and could not be successfully transformed. As such, the results from the DIA methods were compared with the visual estimates using Spearman's rank correlations, to identify any relationships between software-based digital image analysis (sDIA), estimate-based digital image analysis (eDIA) and visual estimates, as well as their relationships to the sediment-specific biotic index. All correlations were interpreted using the Dancey and Reidy (2007) classifications of correlations; 0.1–0.39 = weak, 0.4–0.69 = moderate, 0.7–0.99 = strong.

RESULTS

Comparison of methods

Software-based DIA and eDIA were strongly and positively correlated with visual estimates ($r_s=0.90$, $p < 0.01$ and

$r_s=0.82$, $p < 0.01$, respectively) (Figure 2). Software-based DIA was also strongly correlated to eDIA ($r_s=0.95$, $p < 0.01$). The mean difference between an individual DIA value and the corresponding (same site and season) visual estimate was 11.5 and 11.3 percentage points (for sDIA and eDIA, respectively), whilst the largest difference was 33 percentage points (37% fine sediment compared with 70% fine sediment).

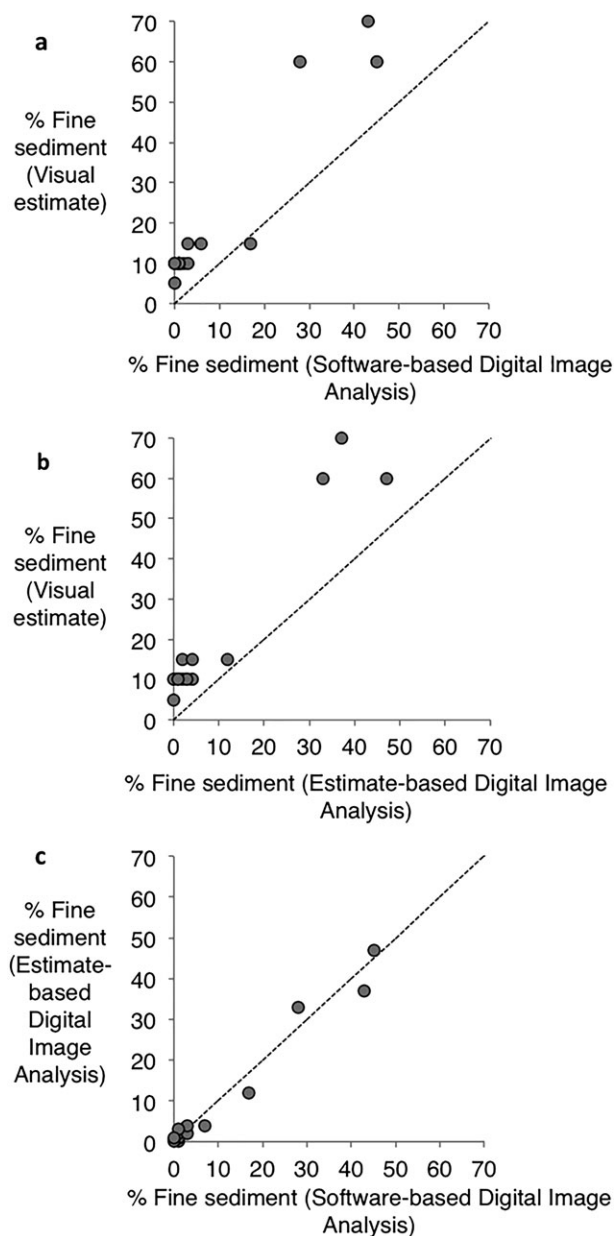


Figure 2. Comparison between (a) visual estimates and software-based digital image analysis (DIA), (b) visual estimates and estimate-based DIA and (c) software-based DIA and estimate-based DIA, at five sites each with three seasonal samples (i.e. $n = 15$)

Precision of software-based and estimate-based digital image analysis

The mean difference between the first operator’s and the independent operator’s sDIA results over the five sites was 1.8 percentage points, with the maximum difference being three percentage points. Similarly, the mean difference between the eDIA from each operator was 1.8 percentage points, with a maximum difference of five percentage points (see Supporting Information).

Fine sediment spatial variation

The variation between transects is illustrated by Figure 3 using data from sDIA. The majority of sites had narrow confidence intervals (<5 percentage points). The exceptions were site 3 (spring) and site 5 (all seasons), with the largest confidence interval being 32 percentage points (site 5—spring).

Biological relevance

All three methods of quantifying fine sediment were strongly negatively correlated to the E-PSI index. The visual estimates and sDIA were similarly strongly correlated to the E-PSI index ($r_s = -0.74$ and $r_s = -0.77$, $p < 0.01$, respectively) with the most strongly correlated method being eDIA ($r_s = -0.87$, $p < 0.01$; Figure 4).

Similar to McHugh and Budy (2005), the visual estimates conducted for this study resulted in higher values for percentage fine sediment, an overestimation of the ‘measured’ method (in their case a hoop-based embeddedness technique). Whilst the differences between DIA and visual estimates may be the result of one method being more accurate than the other, this cannot be tested, as the true value for fine sediment is unquantifiable. The differences between the data from the two methods could also be due to different methodologies, with DIA attempting to represent the percentage of fine sediment across the site using sample patches, and visual estimates summarizing the entire study site. Some of the differences are also likely to be due to the subjectivity of the visual estimate method (Descloux *et al.*, 2010; Roper *et al.*, 2002; Sennatt *et al.*, 2006), as well as the sampling design used in this study for image collection. However, using systematically selected transects and fixed image locations reduces the subjectivity of the technique in comparison with other commonly used methods of quantifying fine sediment. For example, the practicalities of deploying numerous pieces of equipment and/or removing and processing large amounts of sediment mean that many methods result in a limited number of locations being sampled. These locations are often selected based on operator judgements, which may introduce operator error or bias. Techniques that utilize bucket traps, artificial mats, adhesive plates or resuspension techniques often require subjective decisions to be made regarding selection of sampling locations. For example, application of the resuspension technique can require the identification of ‘erosional’ and ‘depositional’ sampling locations (Duerdoth *et al.*, 2015) necessitating a subjective evaluation of stream characteristics, which may vary with flow conditions. Furthermore, it is likely that methods that require such sampling location decisions are influenced by

DISCUSSION

Comparison of methods

The results of this study show that both the sDIA and eDIA methods were strongly correlated with the visual estimates.

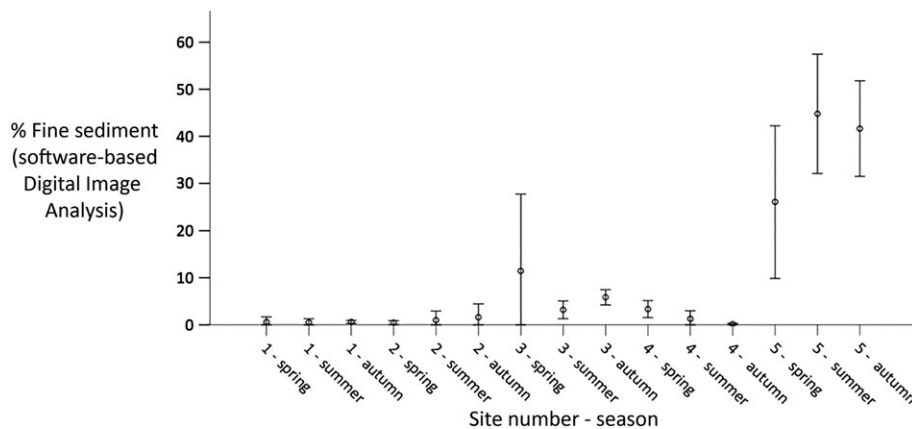


Figure 3. Mean percentage fine sediment values from software-based digital image analysis of five transects (each comprising three images), from five sites [1 (low fine sediment) to 5 (high fine sediment)] over three seasons (spring, summer and autumn). Error bars show 95% confidence intervals

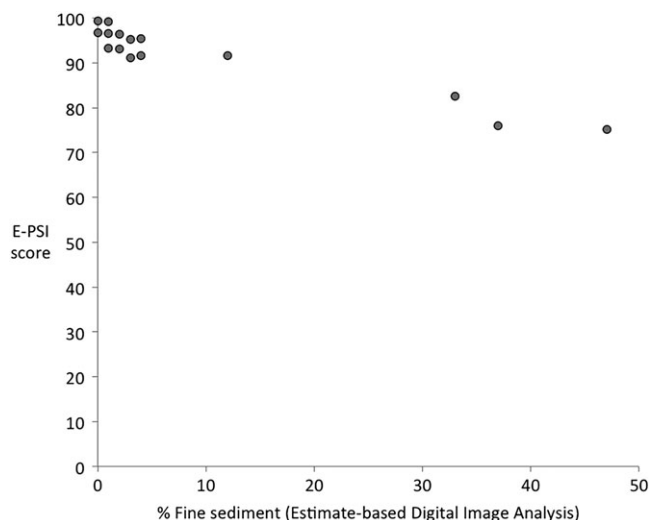


Figure 4. Relationship between % fine sediment (estimate-based digital image analysis) and the Empirically-weighted Proportion of Sediment-sensitive Invertebrates (E-PSI) index

the nature of the substrate or channel itself, with substrates containing large boulders, and/or deeper waters (deeper than sampling equipment), and/or narrow-braided channels (narrower than sampling equipment) being omitted from sampling because of operational constraints. These common limitations impact the ability of methods to represent spatial variation in substrate composition accurately. In contrast, a systematic approach such as that used in this study may more accurately represent spatial variation, although the number of transects/images required to do this is likely to be site specific. Increasing the number of transects/images requires a minimal investment of time, particularly if opting for the estimate-based approach, which requires less than 5 min processing per image (compared with up to 30 min for sDIA).

Digital image analysis is more objective than visual estimates as it allows for particles to be measured and percentages to be computed. The operational limitations associated with DIA are similar to those that are experienced for visual estimates. In conditions where water clarity is poor, the method can prove problematic; however, moving the camera closer to the streambed can, in some cases, overcome this issue. Similarly, low water clarity can also prevent visual estimates. One of the benefits of using digital photography is that photographs can be reviewed instantaneously, something which is not possible for film photography. Other conditions (e.g. low light and rapid flow) leading to poorly lit/focused images require some level of subjectivity in order to identify areas of fine sediment, although digital image contrast can be altered as part of the post-processing to mitigate these problems. The method is restricted to streams that are wadeable, unless a boat mounted system could be used. Nevertheless, one of the significant strengths of the

approach is the ability for images to be archived for verification, therefore providing a means of quality assurance. It is also a non-destructive technique, a characteristic that is particularly desirable when being carried out alongside biological sampling and/or in conservation areas (Naden *et al.*, 2003).

Perhaps unsurprisingly, the sDIA was more strongly correlated to eDIA than to the visual estimates. This is likely to be due to the two methods quantifying fine sediment over the same areas of streambed (captured in the images), whereas the visual estimates consider the site as a whole. This strong correlation ($r_s = 0.95$, $p < 0.01$) suggests that the comparatively rapid approach (up to 5 min per image) of estimating the amount of fine sediment in the images using eDIA could negate the requirement of manually highlighting fine sediment for sDIA, which is a time-intensive procedure (up to 30 min per image). All three methods are likely to be subject to 'fabric errors', which are the result of misclassifications of particle size due to the orientation of particles in relation to the plane of the image, as well as the potential for particles to be partially hidden (Graham *et al.*, 2005b). However, as the focus of the analyses was fine sediment, this error is likely to be minimal.

Precision of software-based and estimate-based digital image analysis

The inter-operator comparison for both methods of DIA yielded relatively small differences between fine sediment values (maximum five percentage points) across the five samples. This is despite the site/season being randomly selected for verification and the independent operator having

no prior knowledge or expectations of the site. Although the precision of visual estimates carried out in this study is not considered, previously published work on this topic has shown the method to have a lower level of precision. For example, a study using 10 different operators showed visual estimates to vary by up to 40 percentage units for sites with the same Wolman counts (Clapcott *et al.*, 2011). However, the operators did not receive any specific training prior to the observations and so this example is likely to be an extreme case. Wang *et al.* (1996) found the precision of visual estimates between six operators to be 'moderate', with confidence intervals of between 5 and 15 percentage points. It is thought that recent standardized training of operators is likely to improve the precision of visual estimates of stream habitat (Poole *et al.*, 1997; Roper and Scarnecchia, 1995). A high level of inter-operator precision was shown for the DIA methods across sites with a range of substrate compositions; however, this analysis involved fewer operators. The sDIA method has a potential advantage over the eDIA method, which introduces some subjectivity to DIA, requiring operators to estimate the percentage of fine sediment in the images (using a 10×10 grid), which is likely to lower its accuracy and precision. Nonetheless, eDIA offers substantial time and cost-savings in comparison with sDIA, requiring approximately one-sixth of the time commitment.

Fine sediment spatial variation

In application, the sampling design used to collect digital images for DIA should be designed to provide suitable representation of fine sediment spatial variation. The use of five transects here is merely an example of how the method could be applied. The relatively narrow confidence intervals for sites 1, 2 and 4 suggest that the sites exhibited little variation in terms of fine sediment coverage and that five transects or less may provide sufficient representation of spatial variation in some situations. The larger confidence intervals for sites 3 and 5 suggest that a greater number of transects/larger area of streambed may need to be sampled to suitably represent fine sediment conditions over certain sites. Environmental factors are likely to influence the number of images required in order to provide an accurate representation of spatial variation. These include the water depth, water clarity and light availability, which limit the area of the streambed captured in each image, potentially necessitating the collection of a greater number of images. Furthermore, sites with a heterogeneous and poorly sorted substrate are likely to require a greater number of images in comparison with more homogenous, well-sorted streambeds, in order to capture images that accurately represent the fine sediment conditions throughout the site.

The positioning of image locations for DIA is not influenced by the presence of boulders, cobbles or bedrock

and so may be able to suitably capture the true characteristics of stream and river substrate. In contrast, techniques such as adhesive plates, artificial mats or resuspension sampling devices are restricted by the diameter of the device and the ability to attach or insert the device into the substrate, although they do benefit from enabling analysis of geochemical properties of the sediment, as part of the post-processing.

Biological relevance

As the true value for the percentage of fine sediment in a stream is unknown, the accuracy of the methods cannot be determined. However, a potentially more meaningful measure (depending on the intended application) is its biological relevance. The E-PSI index was strongly correlated to all three methods of quantifying deposited fine sediment. These results suggest that all three methods have some degree of biological relevance, particularly given the biological basis of the index, which provides a mechanistic linkage for the observed correlations. These strong correlations were observed at systematically selected sites that greatly varied in their environmental characteristics, suggesting that the techniques are indicative of fine sediment conditions across a range of different temperate rivers and stream types. Although the DIA approach does not consider the quality of fine sediment (geochemical and particle size distribution), the percentage cover of fine sediment is likely to be biologically relevant as it relates to niche theory and habitat suitability (Hirzel and Le Lay, 2008) throughout the reach.

Potential for future application

Whilst visual estimates of fine sediment have been found to be some of the most correlated metrics to both land use and invertebrate biotic indices (Sutherland *et al.*, 2010, 2012), the subjectivity of these methods have the potential to result in incorrect conclusions as to the sediment conditions at a site. DIA presents an opportunity to reduce the subjectivity involved in characterizing streambed fine sediment conditions. Such an approach is highly desirable for monitoring and research applications, as well as for river restoration and management projects, which require non-destructive, reliable and ecologically meaningful habitat indicators (Woolsey *et al.*, 2007). Given the budget constraints often placed on these types of application, the eDIA approach may provide a more suitable means of characterizing fine sediment conditions. Further work should be conducted to determine the optimum number of transects or area of streambed that is necessary to provide a good representation of fine sediment conditions across sites with differing environmental characteristics.

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