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# Tracing organic matter sources in riverine suspended sediment: Implications for fine sediment transfers

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#### Abstract

The study of sediment transport in riverine systems has historically focussed on the inorganic fluxes, although the suspended load is known to incorporate both inorganic and organic material. Research on freshwater flocculation indicates that sediment morphology and behaviour depend on both components, as biologic material can alter the hydrodynamic and charge properties of composite particles, which has implications for fine sediment transport and storage in stream systems. This study evaluates both the supply and the quality of the organic matter comprising the suspended sediment in a salmon-bearing stream in northern British Columbia, Canada. Seasonally changing hydrodynamic and biologic conditions are considered in the context of suspended sediment (seston) size. Seston samples were collected in an event-based design for analysis of organic matter sources, suspended particulate, inorganic and organic matter concentrations (SPM, SIM and SOM) and effective particle size distributions (EPSD). Organic matter sources were assessed using carbon: nitrogen (C:N) ratios and the associated stable isotopic signals. Samples and environmental measurements were taken during spring melt, pre-spawn low flows, active salmon spawning, post-spawn, and select rainstorms. SPM and SIM were elevated during active salmon spawning compared to that expected for the given hydrologic conditions. Seasonal patterns of C:N ratios and stable isotopes indicate that organic matter types can be differentiated in seston samples and these sources of organic material exhibit changing dominance in the seston over time. The isotopic signature of seston increases in salmon-derived nutrients when live and decaying salmon are present within the study stream. The largest suspended particles are observed during the period of active spawning when both live and rotting salmon modify instream conditions. This appears to be a function of the combined influence of high SIM, low shear stress, and low C:N ratios. The generation of increased floc, or seston size is of ecological importance in this stream and riverine systems elsewhere as it regulates the freshwater transfers of locally derived nutrients and sediments out of the fluvial system.

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### 1. Introduction

Sediment researchers working in a range of disciplines and environments traditionally measure the chemically dispersed (i.e., disaggregated) mineral fraction to characterize particle size distributions (Droppo, 2001). For questions involving the transfer and storage of fine sediments, such as silt and clay ( $< 63 \,\mu m$ ), which are by nature cohesive, this approach is inappropriate as they are known to move in both marine and freshwater systems as composite or aggregated particles (Kranck et al., 1993; Droppo and Ongley, 1994; Petticrew, 1996; de

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Boer, 1997; Phillips and Walling, 1999). The emphasis in fine sediment aggregation, or flocculation, research has been to characterize the altered composite particle structure (size, shape, density) and behaviour (settling velocity) to determine their influence on the transport and storage of fine particulate material within watersheds. As these fine sediments are vectors of both nutrients and contaminants, increased amounts and periods of storage are significant in the context of habitat conditions and geochemical transfers.

A variant of inorganic sediment is seston, which is a term used in aquatic biology to represent material suspended in an aqueous system, composed of living (e.g., bacteria and algae) and nonliving (e.g., organic detritus and inorganic sediment) fine (0.45 to 1000 µm diameter) particulates (Wallace and Grubaugh, 1996). Flocculation occurs when particles comprising seston collide incidentally due to turbulent flow or gravitational settling with adequate force and favourable chemical and biological conditions. Although hydrodynamic conditions (e.g., shear stress; Milligan and Hill, 1998) are integral to particle collision, and the chemical nature of the particles (e.g., charge) and fluid medium (e.g., conductivity) typically determine combining efficiency in marine systems, there is mounting evidence that biological considerations are significant in freshwater systems (Droppo, 2001; Petticrew and Arocena, 2003; McConnachie and Petticrew, 2004; Leppard and Droppo, 2005). If biological composition is an important regulatory factor in freshwater floc formation and morphology, the annual cycle of changing organic source material to streams and the implications of variable organic matter (OM) quality may be reflected in the changing morphology and transport behaviour of fine sediment.

The ratio of carbon to nitrogen in organic matter has been used to differentiate source material as well as to characterize seston quality (Bouillon et al., 2000). Lower ratios of C:N imply better quality as the increase of nitrogenous products can be readily used by bacteria. Bacteria and other microorganisms are speculated to be a significant biological component controlling flocculation as they generate microstructures referred to as extracellular polymeric substances (EPS). EPS appear to assist in regulating the shape and size, as well as internal complexity, of flocs (Droppo, 2001), all of which inevitably influence floc density and settling velocity, and, by extension, the nature of suspended sediment transport. As EPS are typically generated by microorganisms, both the quantity and quality of dietary sources (e.g., organic matter) should be important factors in the development of freshwater flocs.

Organic matter supply to rivers and biological quality therein, like inorganic sediment delivery, can be very complex, varying spatially and temporally. Organic material can be delivered from the terrestrial watershed (allochthonous) or generated within the aquatic environment (autochthonous) and can be utilized as fine particulate or dissolved, readily available nutrients. Temporal variability in floc morphology may therefore be due in some part to changes in the sources of inorganic and organic material. Investigation of this idea requires accurate identification of organic sources and an estimate of their incorporation into the sestonic component of aquatic systems.

Stable isotope analysis (SIA) has been used successfully by ecologists to identify organic matter sources, as well as to trace trophic pathways (Peterson and Fry, 1987), in marine (e.g., Cifuentes et al., 1988) and freshwater (e.g., France, 1995) systems. To discern the mixture of organic matter sources of a system or secondary consumer, isotopic (C, N) values are determined for representative sources, termed endmembers. Isotopic values of samples representing mixed sources are analyzed and the proportional contributions of endmembers are assessed through mixing models (Phillips and Gregg, 2001). Linear mixing models are used for this purpose to examine two source, single isotope, or three source, dual isotope signatures. Typically, the mixture is some secondary consumer or, in the case of riverine systems, periphyton. Few studies have attempted using a seston mixture to determine the organic matter sources in suspended sediment.

The application of SIA is particularly useful in systems utilized by anadromous fishes (those that move between marine and freshwater habitats) that are known to greatly influence the nutrient supply of freshwater systems for the short time that they are present to spawn (Finney et al., 2000). Specifically, the nutrients produced from decomposing post-reproductive salmon carcasses act to fertilize both the aquatic and riparian environments of freshwater systems (Bilby et al., 1996; Ben-David et al., 1998). This form of organic matter appears to be of higher quality than that derived from terrestrial sources because the latter is more refractory or difficult for microbes to process quickly. Salmonid fishes contribute significantly to freshwater nitrogen budgets. In fact, carcass-derived nitrogen is known to enhance microbial and algal growth (Wold and Hershey, 1999) and should, in turn, lead to greater EPS production potentially modifying floc morphology and transport.

The objectives of this study are to (1) determine if the elemental and isotopic C and N signatures of autochthonous and allochthonous source material and sestonic organic matter can be statistically differentiated in order to (2) establish if seston reflects temporal variation in sources of organic matter to watersheds. If these two objectives are met, the final goal is to (3) determine if temporal changes in the dominant organic matter source within a relatively undisturbed, productive headwater stream results in a corresponding change in the morphology of the suspended sediment.

### 2. Materials and methods

### 2.1. Field site and sampling design

The study watershed is located in the Hogem Range of the Omenica Mountains in the Takla Lake region of northern British Columbia (Fig. 1), an area where research regarding the relationship between forestry activities and the productivity of aquatic ecosystems in British Columbia's central interior has been undertaken (Stuart-Takla Fisheries–Forestry Interaction Project; Macdonald et al., 1992). The O'Ne-eil drainage basin is a small (75km<sup>2</sup>), relatively undisturbed (one access road built in 1980) system at the most northerly extent (55°N, 125°50′W) of the Fraser River catchment in British Columbia, Canada. The basin is part of the Engelmann Spruce Sub-alpine Fir (ESSF) biogeoclimatic zone. The geology of the basin is predominantly argillite and limestone while the surficial material is comprised of glacial tills at higher elevation and finegrained glaciolacustrine sediment in the lowland areas (Ryder, 1995). The main channel is approximately 20km in length with the lower 2km exhibiting excellent conditions for sockeye salmon (*Oncorhynchus nerka*) spawning (Macdonald et al., 1992). The Canadian Department of Fisheries and Oceans has measured stage height in this stream since the early 1990s while annual escapements (adult returns) of these early Stuart salmon stocks to O'Ne-eil Creek have been well documented since the 1950s. O'Ne-eil Creek has an annual escapement range of between 1000 and 50,000 salmon returning between late July and mid-August each year (Poirier, 2003).

A 10m riffle in a straight reach of the main channel, approximately 1500m upstream of the mouth, was sampled during the period of 18 May to 21 August 2001. Samples were collected to evaluate the seasonal changes in fine sediment composition and morphology over a range of hydrologic and biologic events (Fig. 2A). Hydrologic events included (i) the rising limb of the spring melt freshet, (ii) the period after spring melt when discharge declines toward low flows, here called pre-spawn and (iii) five rain events sampled at a range of times relative to the start of each storm (displayed as arrows in Fig. 2A). Biologically important periods were (a) active spawning upon the return of migrating salmon (return date 21 July), and (b) the post-spawn die-off. The spawn



Fig. 1. Map of the Stuart-Takla region of northern British Columbia. The watersheds delineated were instrumented as part of the Stuart-Takla Fisheries–Forestry Interaction Project. Note O'Ne-eil watershed in the center. This map is modified from Macdonald et al. (2003).



Fig. 2. (A) Discharge and precipitation over the 2001 field season for O'Ne-eil Creek in northern British Columbia, Canada. Hydrologic and biologically important periods are identified with shading, while sampled rain events are noted with arrows. (B) SPM and SOM (mg L<sup>-1</sup>) values from triplicate samples of suspended sediment (seston). Error bars represent  $\pm 2$  standard errors (SE). Note scale change between SPM and SOM concentrations. (C) C:N ratios calculated from duplicate samples of filtered seston. Error bars represent  $\pm 2$  SE. (D)  $\delta^{13}$ C and  $\delta^{15}$ N found from the same duplicate seston filters as the C:N ratios. Error bars represent  $\pm 2$  SE.

regime includes the period of active digging of redds (nests for eggs) as well as the start of post-reproductive die-off, while post-spawn is devoid of live fish but incorporates the continued decaying fish carcasses. Spawning salmon are responsible for a considerable amount of streambed modification generally (Soulsby et al., 2001) and specifically in O'Ne-eil Creek where Poirier (2003) has quantified the vertical disturbance of the gravel bed associated with redd construction. This activity contributes significantly to the immediate suspended sediment concentration as the fish resuspend gravel stored sediment, sand size and smaller, cleaning the gravels and promoting high quality incubation conditions for their eggs. During this study, 13,580 sockeye salmon returned to the lower reaches of O'Ne-eil Creek to spawn with an approximate density of 0.7 fish  $m^{-2}$ .

### 2.2. Field sampling

Multiple large mouth bottle samples of water and seston were removed at the thalweg for (i) suspended particulate, inorganic and organic matter concentrations (SPM, SIM and SOM), (ii) particulate carbon and nitrogen content, and (iii) stable isotope analysis ( $\delta^{13}$ C and  $\delta^{15}$ N). Composite 20L samples were collected for effective (in situ) particle size distributions (EPSD). Samples were withdrawn near the water's surface and thus do not reflect depth integrated values. Temperature and conductivity measurements were taken during sample collection.

Velocity profiles were used to estimate shear stresses in the water column (c.f. McConnachie, 2003) and the shear velocity ( $V^*$ ; m s<sup>-1</sup>) along a hydraulically rough streambed was determined using the methods of Gordon et al. (1992). Bottom shear stress ( $\tau_0$ ; N m<sup>-2</sup>) was calculated from data for each sample date as it is directly proportional to the square of the shear velocity and the density of water ( $\rho$ ; kg m<sup>-3</sup>), which is dependent on temperature (Nowell and Jumars, 1984).

Effective particle size distributions (EPSD) of the seston were calculated from analysis of images obtained using a Plexiglas settling tube (c.f. Petticrew and Droppo, 2000) and a charge-coupled device (CCD). An image analysis program, Northern Eclipse (Empix Imaging, Mississauga, ON, Canada) was used to capture sequential images of particles as they settled due to gravity. Dimensions (e.g., diameter, area, perimeter, and shape) of 500–1500 particles for each sample date were measured and recorded using the Northern Eclipse package. Details of the settling technique set-up and imaging resolution are presented in McConnachie (2003).

### 2.3. Suspended sediment filtering

Water samples were filtered through triplicate precombusted and pre-weighed 47 mm diameter,  $0.7 \mu m$ pore size glass-fiber filters for gravimetric determination of SPM. Ashing of the filters at 500 °C and reweighing allowed for estimates of SIM and SOM. A second set of these filters was freeze-dried and analyzed for carbon and nitrogen content as well as stable isotopes (C, N).

### 2.4. Stable isotopes of carbon and nitrogen

Stable isotope mass spectrometry (University of British Columbia, Oceanographic Stable Isotope Labo-

ratory) was used to characterize seasonal sources of organic matter. The isotope ratios for both organic source tissue and suspended sediment filters were measured and expressed relative to conventional standards as  $\delta$  values defined as:

$$\delta X(\%) = (R_{\rm sa}/R_{\rm std} - 1) \times 1000 \tag{1.0}$$

where X is  ${}^{13}$ C or  ${}^{15}$ N determined as parts per thousand (‰),  $R_{sa}$  is the isotopic ratio of the sample (either <sup>13</sup>C/  $^{12}$ C or  $^{315}$ N/ $^{14}$ N), and  $R_{std}$  is the isotopic ratio of the standard (PeeDee Belemnite for carbon and air for nitrogen). The technique enables assessment of seasonal distribution of organic matter sources comprising seston by comparing isotopic ratios from source material with those from filtered suspended sediment samples. Carbon and nitrogen content were measured prior to stable isotopes and C:N ratios were calculated from the resulting values. This more standard ratio is often used to evaluate sources of organic matter in aquatic systems because autochthonous (instream productivity) material exhibits much lower values (<15) than allochthonous (terrestrially derived) organic matter (Owen et al., 1999). Statistical analyses of seasonal isotopic patterns were performed using Statistica 6.0 (Statsoft, 1998). The Shapiro-Wilk's test was used to assess normality of variables and the Kruskal-Wallis non-parametric analvsis of variance was employed for detection of significant differences.

Tissue from terrestrial vegetation comprising spruce needles, willow leaves, alder leaves, and birch leaves, algae, periphyton, and salmon flesh was collected and stored in 1.2mL centrifuge tubes and subsequently freeze-dried. Isotopes of carbon (<sup>13</sup>C) and nitrogen  $(^{15}N)$ , as well as the  $\delta$  values of each, were determined. The average C:N ratio for each source material was obtained from at least 3 replicates taken over the field season. Algal matter consisted of free-floating assemblages, and samples were collected from blooms growing in areas of slow-moving water within the reach. Algal samples were collected in August only due to an apparent lack of availability in the stream at any other time. Periphyton was found attached to large woody debris and other streambed substrates. Periphyton samples were collected over the season, but were not used as an organic source population in the analysis because this organism potentially utilized nutrients from their organic substrates as well as from the water column, which connotes that its isotopic signature is comprised of a mixture of endmembers. Furthermore, periphyton was collected by a scraping method, which made it difficult to ensure that

Table 1 Averaged C:N ratios for different types of organic material sampled in or adjacent to O'Ne-eil Creek

Source type	Tissue origin	N	Mean	2 SE
Allochthonous	Willow	4	18.38	2.47
Allochthonous	Spruce	4	41.91	14.84
Allochthonous	Alder	4	28.11	2.62
Allochthonous	Birch	2	20.06	3.26
Autochthonous	Periphyton	4	8.33	2.43
Autochthonous	Algae	3	9.23	0.42
Autochthonous	Salmon	4	3.41	0.13

Allochthonous (terrestrially derived) and autochthonous (produced in stream) organic material are characterized by a C:N ratio of above and below 15, respectively.

samples were not contaminated by fragments of the attachment surface. Phillips and Gregg (2001) suggest that, for modeling purposes, samples from the three source (i.e., endmember) populations should remain independent so as not to complicate the results. For this reason, periphyton was excluded from further analysis.

A dual isotope (C, N), three endmember (algae, salmon, and terrestrial vegetation) mixing model based on mass balance equations (Phillips and Gregg, 2001) was used to quantitatively characterize seasonal trends in source partitioning. The spreadsheet used to determine source proportions, variances, standard errors (SE), and confidence intervals can be accessed at http://www.epa.gov/wed/pages/models.htm.

# 3. Results

### 3.1. SPM and SOM

As expected, the SPM and SOM increase on the rising limb of the snowmelt flood and decrease on the falling limbs of the large floods (Fig. 2B). The notable exception is a relatively large increase in concentrations of both SPM and SOM during the spawning period that is characterized by low flows.

### 3.2. Organic matter source composition

Table 1 provides the results of C:N ratios for the range of allochthonous vegetation and autochthonous organisms sampled for endmember characterization. The highest value was for spruce needles  $(41.91 \pm 14.84)$  and the lowest of  $3.41 \pm 0.13$  was for salmon flesh. Owen et al. (1999) found a C:N ratio of 15 to be the threshold between allochthonous (>15) and autochthonous material. The data displayed in Table 1 fit this model, as the instream periphyton, algae and salmon are all significantly lower than 15, while all types of riparian vegetation analyzed exceed this value.

Fig. 3 shows the measured endmember isotope signatures and a mixing model result for a hypothetical seston sample. The proportional source material composition for the seston mixture is determined by



Fig. 3. Hypothetical partitioning of source contributions from three selected endmembers (terrestrial vegetation, algae, and salmon flesh) for stable isotope mixtures collected as filtered seston. The approximate contribution from terrestrial vegetation, salmon, and algae is 49%, 24%, and 27%, respectively. Error bars represent  $\pm 2$  SE.



Fig. 4. (A) Percentage contribution from the three endmember sources to seston as determined from a linear, dual isotope (C, N) mixing model. Averages are shown  $\pm 2$  SE for four event periods comprising spring melt, pre-spawn, spawn, and post-spawn (N=5 for each). (B) Percentage contribution from the three endmember sources to seston as determined from a linear, dual isotope (C, N) mixing model. Averages are shown  $\pm 2$  SE for duplicate samples collected during each of five rain events over the 2001 field season. July rain dates represent pre-spawn, Aug 2 and Aug 3 storms occurred during the spawn period while the Aug 21 rain event reflects post-spawn conditions.

partitioning the three source types presented in Fig. 3. The measured  $\delta^{13}$ C and  $\delta^{15}$ N signatures, and two standard errors, are plotted for each of terrestrial vegetation, algae, and salmon, as well as for the hypothetical sestonic mixture which falls within the triangle formed by the three endmember values. The expectation is that its position within the triangle would vary seasonally due to proportional changes in source contributions.

### 3.3. Sestonic organic matter composition

Fig. 2C depicts the seasonal pattern of the C:N ratio derived from suspended sediment samples taken throughout the season. C:N ratios are high, being >15, from spring to mid-July, prior to salmon migration into the study reach. The ratios decrease when salmon enter the system, and continue to decline during the salmon die-off, when post-reproductive carcasses rot within the stream. For all but one of the samples, the ratios are below 15. This elevated sample is associated with the falling limb of a storm event. The temporal display of stable isotope data for the same seston samples exhibits a changing seasonal trend (Fig. 2D). The Kruskal-Wallis non-parametric assessment of variance was used, due to the limited sample size, to detect differences between event periods. The carbon data establish a slight, but not statistically significant, enrichment of the heavier isotope from spring melt to pre-spawn. The average values ( $\pm 2$  SE) for these event periods are  $-26.82\pm$ 0.12% and  $-26.57\pm0.14\%$ , respectively. The enrichment is more pronounced, and statistically significant (p=0.002), when salmon enter the reach  $(-26.16\pm$ 0.22‰ as averaged for the spawn period). The peak individual value (-25.42±0.18‰ from duplicate samples) occurs on August 13th just at the end of the active spawn period when instream conditions are characterized by a few living, reproducing salmon mingled with in situ rotting of earlier returnees. The isotopic carbon values for the post-spawn period exhibit a mean of - $25.62\pm0.14\%$  at this time when only dead, decaying carcasses are present in the stream. The nitrogen isotope follows a similar enrichment pattern with the lowest values of 2.02±0.22‰ observed at spring melt and highest mean values of 5.60±0.36‰ sampled during post-spawn. Isotopic N is significantly higher (p < 0.001) for periods when living and decaying fish are in the stream as compared to when fish are absent (i.e., spring melt and pre-spawn). The increase in isotopic N in the seston samples is evident in the non-averaged values (Fig. 2D).

Daily duplicate filtered seston samples were grouped for each of the five sampling regimes and run through the dual isotope, three endmember mixing model developed by Phillips and Gregg (2001) to determine proportional organic matter contributions for each period. The model compares the isotopic values of  $\delta^{13}$ C and  $\delta^{15}$ N measured in seston samples to isotopic signals measured from the source material tissue samples identified in Table 1. Because the model used was based on linear extrapolation, and the sample size was limited, the percentage of terrestrial and algal contribution exceeds 100% and falls below 0%, respectively, on some sample dates (Fig. 4A and B). Overall, the results indicate that the predominant



Fig. 5. Event averages ( $\pm 2$  SE) of  $D_{99}$ , C:N ratio and suspended inorganic matter (SIM) for each event period category of spring melt, pre-spawn, rain, spawn, and post-spawn. The maximum shear stress calculated for each period is shown.  $D_{99}$  values were extracted from effective particle size distributions (EPSD).

source of organic matter to the seston load changed over the season (Fig. 4A). For the spring melt period, the model suggests that all organic material is derived from terrestrial vegetation, while the salmon and algal sources contribute minimally. General trends indicate a significantly reduced contribution from terrestrial sources over the sampling period. For these data spring melt and prespawn periods do not differ from each other, but they do differ from the post-spawn period. The spawn average is not statistically different from any of the other event periods due to the variability exhibited in individual samples over that event period (see Fig. 2D). The contribution from salmon increases during active spawn (32.8 $\pm$ 14%) and by post-spawn the riparian vegetation and salmon appear to represent an equal proportion of the sestonic organic matter of approximately 46% each. At this time the algal contribution becomes positive and represents the remaining 7%. The general trend is for the terrestrial organic sources to reduce from spring melt to post-spawn while the percentage contribution to sestonic organic matter of salmon increases. Average values of spring melt, pre-spawn, and spawn are not different from each other nor are spawn and post-spawn, but post-spawn differs from spring melt and pre-spawn. While there are no statistical differences noted in the algal response over the season the only positive contribution to the seston is noted in post-spawn.

The rain events, which were not included in the foregoing analysis, can be used to corroborate this seasonal pattern of changing sestonic composition. Fig. 4B shows the results of the mixing model for each of the five rain events. A comparison between the two storms occurring pre-spawn (July 7th and 9th), and the storms associated with salmon spawn (August 2nd and 3rd), indicates a shift from a dominant contribution of terrestrial sources toward a mixture of riparian and instream organic matter types over time. The last rainstorm during the post-spawn period (August 21st) indicates a decline in salmon influence and an increase in terrestrial sources once again.

### 3.4. Organic matter influence on particle size

Fig. 5 depicts the changing trend of aggregated particle size  $(D_{99})$  with C:N, maximum shear stress (for the respective event period), and suspended inorganic matter. Larger average in situ particle sizes were found from EPSD for the rain and spawn categories, while spring melt, pre-spawn, and post-spawn consisted of particle sizes that were statistically the same. Spring melt is characterized by relatively high discharge (Fig. 2A), SIM, and C: N, while low flows and lower C: N values, combined with relatively high SIM, occurred during the spawn period. The seasonal trend is that of decreasing flows and C: N ratios over time, with SIM being elevated during high flows and the spawn period. Maximum shear stress for the spring melt category was much greater than that of the spawn with values of 60.2 and 9.0 N m<sup>-2</sup>, respectively. Overall, larger particle sizes are associated with higher quality organic matter sources (low C:N), low shear, and available inorganic sediment.

### 4. Discussion

Aquatic ecologists, and some geomorphologists who have adopted a more ecological approach to studying rivers (Olley, 2002; Peart, 2003; Thoms, 2003), are predominantly concerned with the flux of energy and matter through watersheds. This requires knowledge of the dynamics of organic matter through the various trophic levels in aquatic systems. Organic matter supply to rivers, like inorganic sediment delivery, can be very complex varying temporally and spatially but as well in quality. Riverine organic material can be delivered from the terrestrial watershed (allochthonous) or generated within the aquatic environment (autochthonous) and as well can be utilized as fine particulate or dissolved, readily available nutrients. It is evident that stream systems are not only energetically modified in a geomorphic context, but are also bioenergetically dynamic. Flux investigations of organic matter and nutrients share similar conceptual approaches to that of inorganic sediment as they are influenced by a number of the same environmental processes that regulate sediment delivery and storage.

In undisturbed temperate rivers, the inorganic and organic fluxes are generated by seasonal shifts in both the hydrologic conditions and contributions from different organic sources. High discharges during spring melt and storm events are associated with high inorganic sediment loads derived from channel bed and banks and potentially terrestrial soil erosion. Low flows carry decreased loads of mineral material. In this region of northern British Columbia, leaf litter from riparian vegetation is delivered in small amounts throughout the year, but in September the major annual leaf fall occurs. Litter is delivered directly to the stream and to the floodplain where it decays over winter. During snowmelt breakdown products of litter are available for transport to the stream channel. A very rough estimate of riparian contributions to the floodplain upstream of the sample reach suggest that 9680kg of deciduous and 760kg of coniferous litter were delivered the previous autumn.

In these salmon-bearing streams the sockeye return over a three to four week period (late July to late August), select mates and suitable nest sites and spawn over a period of days. Their death a few days after reproduction is followed by carcass decay which generates a major pulse of autochthonous organic breakdown products within the stream. Due to the extended period over which the early Stuart salmon return to their natal streams this die-back process starts to overlap with active spawning within one or two weeks after spawning begins. While the die-off continues through to the end of August when live spawners are no longer in evidence, the bulk of the breakdown products can be delivered during active spawning. The calculated salmon biomass decaying upstream and within this specific study reach in 2001 was estimated at 12,960kg. Note that this is approximately the same order of magnitude as the estimated terrestrial contribution.

Instream blooms of algae and periphyton are dependant upon nutrient supply, light and temperature. Chlorophyll a maximums, reflecting these populations, in the Stuart-Takla streams have been observed during the pulse of nutrients associated with salmon die-back (Johnston et al., 2004). McConnachie (2003) measured post-spawn chlorophyll a at  $0.47\pm0.08$  mg m<sup>-3</sup>, which if cumulated over a 10 day post-spawn period would represent an algal contribution of 0.81 kg. While this is not an annual value which can be directly compared to the other endmember biomasses, this estimate is for the period when algal biomass is at its maximum in this system and it is considerably smaller than the other estimates.

The present study is a reach-scale investigation of the influence of seasonal changes in organic matter sources to the properties of seston in a highly productive, salmon-bearing system. The sample design allows a comparison of hydrologically significant events (e.g., spring melt and summer storms) to low flow periods, while the importance of the biologic events (spawn and post-spawn) can be differentiated via a comparison to pre-spawn conditions. Here we have used elemental and isotopic (C, N) analysis to identify the seasonal pattern of organic matter source types to stream seston.

# 4.1. Carbon and nitrogen signatures of organic source materials and seston

As indicated the dominant sources of organic matter to O'Ne-eil Creek were identified as terrestrial riparian vegetation and returning stocks of spawning sockeye salmon. Other instream sources included algal and periphyton growth over the open water season. The C: N ratio for allochthonous and autochthonous source materials separate markedly at a value of 15 (Table 1) and as well the isotopic signatures for the three dominant source materials are statistically distinct (Fig. 3), which enables differentiation between sample mixtures. The results determined for O'Ne-eil Creek autochthonous and allochthonous organic sources (Fig. 3) are similar to those of Ben-David et al. (1998), who reported that spawning Pacific salmon exhibit higher proportions of heavier carbon and nitrogen isotopes ( $\delta^{13}C = -18.65 \pm$ 0.18‰ and  $\delta^{15}N=13.01\pm0.13\%$ ) than terrestrial plants (means of  $\delta^{13}$ C  $\approx -27\%$  and  $\delta^{15}$ N  $\approx 0\%$ ; France, 1997).

Elemental C:N ratios can be analytically detected in stream seston (Fig. 2C) and indicate that results from replicate filtered sestonic samples allow statistical differentiation over the sampling period. As well, isotopic C and N results shown in Fig. 2D denote that averaged values of filtered residue from replicate instream seston samples exhibit standard errors, which allow statistical discrimination of their organic matter signature. Given that changes in organic matter signatures can be differentiated in stream seston, and that the signatures of the dominant organic source materials to O'Ne-eil Creek are distinct, the second objective of this study, to determine if the seston signatures reflected the changing temporal contribution of source material to the stream system, was addressed.

# 4.2. Temporal variation in sestonic source composition

The decreasing value of the C:N ratio in seston over the sampling period reflects the seasonal change in organic matter availability (Fig. 2C) as the late July reduction to values below 15 is associated with the arrival of salmon within the study reach. During the period of fish spawning and die-off the seston exhibits a lower elemental signal although the ratio does not decline to that measured for salmon flesh (Table 1). However, the addition of abundant fish decay products with elevated nitrogen contributions (Ben-David et al., 1998; Johnston et al., 2004) acts to reduce the C:N ratio of the creeks seston.

Variation in the C:N ratio is also influenced by hydrological events occurring in the system. A single

date in Fig. 2C that does not comply with the rest of the samples collected during the spawn period is associated with the falling limb of a storm event. Resuspension of material stored in the streambed, and the potential introduction of more terrestrial organic material from streambanks and riparian areas are expected to alter the C:N ratio at these times. Source changes such as these were expected during rainstorm events and it would be of interest to collect future samples over the full storm hydrograph to determine if source differences reflect changing contributing areas on the rising and falling limb of storms.

Stable isotopes provide an even more detailed means of characterizing organic matter sources. The data presented in Fig. 2D indicate little temporal variation in  $\delta^{13}$ C (-27‰ to -26‰) and  $\delta^{15}$ N (2‰ to 3‰) until the salmon enter the reach. At this point, a steep and statistically significant increase in the heavier isotopes of each element occurs; steeper for nitrogen than carbon. Then, when live salmon are no longer present in the stream, both carbon and nitrogen isotopes in the seston begin to decline; at this time the last pulse of decay products are being utilized by instream algae (Fig. 4A) and flushed downstream.

Dual isotope, three endmember modeling (Fig. 4A and B) corroborates the visual stable isotope pattern (Fig. 2D) indicating the technique's usefulness in differentiating source material in seston over the season. Dominant terrestrial proportions in spring melt and prespawn seston reflect the availability of floodplain and channel stored detrital leaf litter, while low values of sestonic salmon and algal signals are consistent with their scarcity in the system at that time. Seston sampled during the spawn stage, which includes part of the carcass decay period, exhibits a decrease in terrestrial composition as the instream pulse of salmon-derived nutrients incorporate into the streams particulate load. The post-spawn seston exhibits significantly less terrestrial organic matter and increased salmon and algal signals reflecting instream conditions at the time. The general trend of decreasing allochtonous and increasing autochthonous proportions of nutrients in seston over the summer season is also observed in the stable isotopes measured for the rain events (Fig. 4B). However, less distinct changes in both the proportion of salmon and terrestrial organics are noted over time due potentially to smaller numbers of replicates and because of resuspension and influx of terrestrial material as noted previously. The final sample date, the August 21 rain event, shows an increased terrestrial input, but as the substantial contribution of leaf litter associated with autumnal scenescence of deciduous trees had not vet

23

begun, we surmise the allochthonous contribution was delivered from floodplain or channel storage.

### 4.3. Variation in organic matter source determinations

Phillips and Gregg (2001) performed a sensitivity analysis of this linear mass balance model and found that large differences in isotopic signal between sources reduced the error (i.e., doubling the difference reduced the uncertainty by half). Further, sample size is important when dealing with source samples exhibiting similar isotopic signatures. Thus, increasing the number of samples collected, especially for terrestrial vegetation, which varies significantly in terms of species and season (France, 1995), should improve the resolution of this model. In this case, the isotopic signatures of all terrestrial source material sampled (Table 1) was used to generate a mean and standard error for the mixing model endmember. The collection of tissue samples at set intervals over the season and application of the model to sources collected specifically at the same time as the mixture should also improve investigation of natural patterns.

The rain event sampled on August 3rd exhibited an elevated C: N ratio, discussed earlier, but note in Fig. 2B the large variation in gravimetric measurement of suspended organic matter (SOM). This same variability was not apparent in the estimates of elemental ratios or isotopic carbon and nitrogen. This implies that while the quantity of organic material was highly variable on that sample date, the quality or composition of it changed very little. Large variation in C:N ratios does occur on several dates in the sampling period, likely indicating short term changes in material collected in surface grab samples. The variation in characterizing sestonic quality analysis could potentially be reduced by collection of larger bulk samples of seston, which integrate a longer (i.e., minutes versus seconds) time period which could then be sub-sampled for replicate analysis.

# 4.4. Regulation of floc morphology

### 4.4.1. Suspended inorganic matter

Given a constant sediment supply, conventional hydraulic theory predicts an increase in suspended sediment concentration with flow because increased turbulence and shear stress enhance the entrainment and transport of sediment. The source and supply of available sediment, however, does change as a function of factors such as the season and antecedent flow conditions. Spring melt is characterized by flushing of a large pulse of stored inorganic and organic material from both the channel and the floodplain. This includes scouring of stream banks and removal of debris dams. Rains events are presumed to draw from similar source supplies, although response is dependent on discharge and seasonal conditions. In low flows sediment sources are limited predominantly to instream supplies. The data from this study follow the expected pattern of increasing SPM with increased flows (Fig. 2A and B), but deviations are observed during the active salmon spawning period. These observations indicate that salmon spawning activity is associated with an increase in suspended inorganic material.

Fisheries literature (Jones and King, 1950; Jones, 1959), as well as geomorphic investigations into gravel bed conditions (Kondolf and Wolman, 1993; Soulsby et al., 2001; Poirier, 2003), have noted that female fish dig spawning redds by exerting enough force on the bed to move the gravels. After spawning, this same force, in concert with the ambient flow, is used to bury eggs. During this digging process, the gravels are cleaned of smaller material, which is suspended and transported some distance downstream. In a stream reach where hundreds of salmon are spawning simultaneously, a considerable amount of gravel stored fine sediment is suspended. Petticrew (2005) reports similar SPM concentrations measured in active spawn conditions to those measured in the same stream, free of fish, moments after manual gravel disturbance at a site several meters upstream. SPM concentrations are highly variable during the spawn as clouds of fine sediment, aggregated fines and sands are suspended during gravel disturbance, but the sand size and denser aggregated particles tend to settle over a short time and distance. The data in Fig. 5 indicate this biotic resuspension of inorganic sediment when significantly increased SIM concentrations occur during the low discharge, active spawning period. Concentrations return to the expected lower levels associated with the respective discharge when live salmon are no longer present within the reach.

### 4.4.2. Suspended organic matter

A relationship similar to that of SPM is expected for the SOM pattern and the data comply. Spring melt is characterized by elevated organic matter concentrations. A pulse of organic material moves with inorganic material that has been entrained after storage over the winter months. The proportion of organic and inorganic components of the sediment (seston) load is relatively constant over the sampling season with organic matter comprising on average  $27\pm3.4\%$  of the total sample. While the seasonal proportion of organic matter does not vary widely in this system, we have shown that the composition and quality changes over time in O'Ne-eil Creek.

# 4.4.3. Aggregation of inorganic and organic matter in seston

As the supply of both inorganic and organic material is important in regulating freshwater flocculation (Droppo et al., 1997; Woodward et al., 2002), it would seem that, if there is seasonal variation in the fundamental building blocks of aggregated particles, then there should also be temporal changes to the floc morphology (e.g., particle diameter, shape and density). If this is indeed the case, it is probable that the presence of salmon influences particle morphology. We suggest that the activity of spawning salmon alters the supply of both inorganic and organic material to the water column by: (1) increasing the available inorganic sediment to the water column compared to pre-spawn conditions (i.e., in the absence of salmon), and (2) contributing a high quality, marine-derived flux of nutrients to freshwater systems (Fig. 5). The literature has long recognized the role of spawning salmon in resuspending stored particles from the gravels (e.g., Jones and King, 1950; Soulsby et al., 2001), and evidence for larger aggregated structures moving in the water column during these periods of low discharge has been documented (Petticrew, 1996; Petticrew and Droppo, 2000; McConnachie and Petticrew, 2004; Petticrew, 2005). Morphological results for this same 2001 suspended sediment data set presented in McConnachie and Petticrew (2004) indicate that the largest aggregate particles (>1000 µm diameter), which exhibit the lowest densities and settling velocities, occurred predominantly in the spawn period and rain events during spawn. While these low density composite particles are likely to be maintained in suspension even in low flows, the denser, faster settling (>5 mm s<sup>-1</sup>) aggregates also found in the water column during active spawning may not be advected as far thereby returning to storage in, or on, the gravel surface over a short distance (McConnachie, 2003; Petticrew, 2005).

The stable isotope analysis reveals that organic source type is important for aggregate structure in O'Ne-eil Creek. The largest particles were found during the salmon spawn (Fig. 5), a time when the dominant organic source type changes from terrestrial material to higher quality (low C:N), marine-derived organic matter introduced to the freshwater system by the salmon vector. With the presence of fish in the stream both carbon and nitrogen signals changed significantly in terms of the heavier isotopes. With this in mind, we speculate that either (1) the quality of salmon decay products are more conducive to floc-building, or (2) increased nitrogen may have an important role in regulating floc size. This could be either due to the 'sticky' nature of nitrogenous substances or because of the high nutritional value for microorganisms that nitrogen compounds provide (France, 1998; Bouillon et al., 2000). High quality organics lead to increased bacterial activity (Wold and Hershey, 1999), which suggests greater probability for attachments through increased production of extracellular polymeric substances (EPS).

# 4.4.4. Instream shear stresses

In general, floc size and shear stress show an inverse relationship (Fig. 5). This pattern corroborates the laboratory findings of Spicer and Pratsinis (1996) and Milligan and Hill (1998), who observed an inverse relationship between turbulence and maximum floc size, although the range of shear stress was much larger for the former study. Other researchers (e.g., Tsai et al., 1987; Burban et al., 1989) have reported a similar relationship in natural systems. It appears that increasing shear stress in natural systems induces floc breakage, which results in more compact aggregates, and continued breakup and reformation eventually results in much more stable particle structures (Droppo et al., 1997; Petticrew and Droppo, 2000).

In an effort to characterize the changing structure and composition of the fines stored in the gravels of salmonbearing streams, Petticrew and Arocena (2003) collected, sized and chemically analyzed the aggregated sediment collected in gravel infiltration bags in this same reach during pre-spawn through post-spawn of this same sample year (2001). They found that the smallest mass of fine sediment and the smallest aggregate sizes were stored in gravels during mid-spawn. Aggregate density increased during this mid-spawn period, which suggests that the physical activity of the spawning fish modifies not just the amount of gravel stored sediment but also the aggregate structure. The gravel stored aggregates, both before and after live fish and fish carcasses are evident in the stream, were larger in diameter and in greater abundance than when salmon were actively spawning. This implies that local shear stresses, generated by salmon cleaning the gravels of fines, resuspend the larger aggregated particles that had then become available to the water column, as evidenced in Fig. 5. Aggregate particle sizes  $(D_{99})$  measured in the water column during the rain events of 2001 (Fig. 5) are similar in size to the spawn samples, potentially indicating a similar gravel stored source of seston that had become entrained by the increased storm flows. While the local shears of some storms and spawning salmon would break apart the weaker flocs, some aggregates, as shown by the  $D_{99}$ , appear to resist the increased energy environment.

### 4.4.5. Floc morphology summary

There are seasonal patterns in the environmental, chemical, and biological variables that have been shown to influence aggregate structure in O'Ne-eil Creek. Shear stresses, and concentrations of SIM and SOM are regulated by hydrologic processes, and during a portion of the season by the physical activity of spawning salmon. Overall, the data here indicate that the largest aggregate particle sizes are associated with elevated SIM, low C:N ratios, and low shear stresses. The importance of the role of available inorganic and organic matter is emphasized by comparing the spawn and postspawn categories in Fig. 5. Both event periods are characterized by floc-enhancing conditions of low shear and low C:N ratios, but the spawn period has more inorganic sediment and salmon breakdown products available in the water column. The fact that inorganic material is introduced into the water column during a time of increased organic loading and favourably low hydraulic conditions provides suitable conditions for flocculation or increased effective particle size.

The above findings provide evidence of the importance of combined abiotic and biotic contributions of salmon in regulating the delivery of sestonic components to the stream system. The physical resuspension of fine sediment by salmon and the delivery of salmon-derived organic matter to the stream during die-off are associated with both large, slow settling flocs and smaller, denser, faster settling aggregates (McConnachie and Petticrew, 2004). This would result in freshwater transfers of nutrients out of the system via the large, slow settling flocs and the retention of locally derived nutrients and sediments via the settling of the smaller, denser aggregates. While the relatively undisturbed system studied here did not exhibit poor gravel habitat conditions associated with an excessive amount of stored fine sediment (Petticrew and Arocena, 2003), an increased supply of inorganic sediments due to natural or anthropogenic activities in the watershed when salmonderived nutrients are abundant would be expected to induce flocculation, resulting potentially in both inferior incubation habitats and altered nutrient fluxes within the system.

# 5. Conclusions

The data presented provide evidence for the ability of stable isotopes to detect the changing relative contribution of salmon and terrestrial organics in suspended sediment samples and presumably gravel stored fine sediments. The combined analyses of C:N ratios and their stable isotopes strongly indicate that seasonal changes in dominant organic matter sources are important for freshwater flocculation. The presence of salmon in streams is associated with increased particle size within the water column due presumably to the combination of biotic resuspension fine inorganic sediment and introduction of high quality, nitrogenbased, microbial enhancing organics. This process is of ecological importance as it regulates the freshwater transfers of nutrients out of the fluvial system via the large, slow settling flocs and the retention of locally derived nutrients and sediments via the settling of the smaller, denser aggregates.

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