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Hydrologic control of nitrogen removal, storage, and export in a mountain stream

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Abstract

Nutrient cycling and export in streams and rivers should vary with flow regime, yet most studies of stream nutrient transformation do not include hydrologic variability. We used a stable isotope tracer of nitrogen (^{15}N) to measure nitrate (NO_3^-) uptake, storage, and export in a mountain stream, Spring Creek, Idaho, U.S.A. We conducted two tracer tests of 2-week duration during snowmelt and baseflow. Dissolved and particulate forms of ^{15}N were monitored over three seasons to test the hypothesis that stream N cycling would be dominated by export during floods, and storage during low flow. Floods exported more N than during baseflow conditions; however, snowmelt floods had higher than expected demand for NO_3^- because of hyporheic exchange. Residence times of benthic N during both tracer tests were longer than 100 d for ephemeral pools such as benthic algae and wood biofilms. Residence times were much longer in fine detritus, insects, and the particulate N from the hyporheic zone, showing that assimilation and hydrologic storage can be important mechanisms for retaining particulate N. Of the tracer N stored in the stream, the primary form of export was via seston during periods of high flows, produced by summer rainstorms or spring snowmelt the following year. Spring Creek is not necessarily a conduit for nutrients during high flow; hydrologic exchange between the stream and its valley represents an important storage mechanism.

Hydrologic transport by streams and rivers links processes in one region with those much farther away, a notable example being the eutrophication of estuaries and coastal oceans by nutrient sources high in the watershed (Rabalais et al. 2002). Rivers transport materials across the landscape while simultaneously transforming and storing them (Meyer and Likens 1979). Much recent research has examined uptake and storage of nutrients and has shown that streams can rapidly transform nutrients (Peterson et al. 2001; Hall et al. 2009), store them for the short term ($<$ 60 d; Dodds et al. 2000; Tank et al. 2000; Hall et al. 2001) or in the case of nitrogen (N), be an outright sink through denitrification (Royer et al. 2004; Mulholland et al. 2008, 2009).

The balance between how much and how long streams retain elements vs. transport them downstream is central to our understanding of watershed nutrient export (Bernhardt et al. 2005; Roberts and Mulholland 2007; Brookshire et al. 2009). One obvious control on this balance between retention and transport is variation in stream discharge (Doyle 2005). Material transport should dominate during high flows, while material retention should dominate during low flows when streams have higher uptake rates and lower export. This pattern was first documented by budget studies (Meyer and Likens 1979), in which import and export of elements were measured across a range of stream flows. A central finding of budget studies in small streams is that most element export occurs during a small fraction of the time when stream flow is high (Meyer and Likens 1979; Webster et al. 1990). However, these budget approaches do not quantify the processes responsible for transforming or retaining nutrients within stream reaches; stream spiraling techniques are necessary (Newbold et al. 1981; Mulholland et al. 2001). Spiraling methods measure rates of uptake and residence times of dissolved nutrient pools within a reach. Hundreds of stream spiraling measurements show that nutrients are quickly removed by streams (Ensign and Doyle 2006; Tank et al. 2008), yet few of these studies have linked uptake with turnover of particulate nutrient pools because isotope tracers are required to estimate storage and fate of N.

Despite potentially large seasonal variation in stream flows, most N spiraling studies in streams, whether isotope tracer studies (Peterson et al. 2001), or measures of uptake using nutrient addition experiments (Hall and Tank 2003), are intentionally conducted at baseflow, with few exceptions (Valett et al. 1996; Tank et al. 2000; Merriam et al. 2002). Spiraling studies, by themselves, cannot consider the role of hydrologic variability, which is a limitation of the spiraling approach (Fisher et al. 2004). Consequently, we do not know how spiraling rates measured at baseflow change when the stream is flooding. Nor do we know how subsequent spates control storage and export of N that is stored in the benthos. Isotope labeling studies suggest that N pools in streams are resistant to scouring by spates when flows increased 10–20-fold (Tank et al. 2000; Merriam et al. 2002), but the overall residence times of tracer N were short, suggesting that small streams do not store N for long periods (Ashkenas et al. 2004). It is not likely that rates of nutrient uptake are constant across large variations of stream flow because high flows may alter nutrient inputs and uptake processes. For example, nutrient concentration and export are often higher during

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high flow periods of the year (Creed et al. 1996; Baron and Campbell 1997), which may saturate uptake (Mulholland et al. 2008). High flows may scour algae and reduce benthic demand for nutrients. On the other hand, high flows may exchange water and nutrients with floodplains thereby increasing storage.

To consider the role of hydrologic processes in nutrient cycling, it is not sufficient to only examine variation in stream flow; it is also necessary to consider the spatial extent of streams beyond the main channel (Triska et al. 1989; Fisher et al. 1998). Hydrologic exchange between streams and groundwater increases the travel time of water down-gradient, and puts stream water in contact with biofilms attached to alluvial sediments and buried organic matter. This hyporheic exchange may contribute to N retention in many streams (Triska et al. 1989; Holmes et al. 1994, 1996) by fostering denitrification through anoxic flow paths with plentiful organic substrates (Holmes et al. 1996; Baker et al. 2000) or by vegetation uptake (Peterjohn and Correll 1984; Ashkenas et al. 2004). The nature of these surface–groundwater transformations may depend on hydrologic variability.

Here we show how a stream cycles N differently at hydrologic extremes, and then we examine how variation in stream discharge controls the fate of stored N during 1 yr. We hypothesized that N uptake and storage would be highest during baseflow because of high uptake rates and low transport conditions, and that N export would be highest during floods because of high discharge (Doyle 2005). We used a ^{15}N tracer addition of nitrate to measure N uptake, storage, and export for two seasons: during a snowmelt flood, and during baseflow. The stable isotope tracer allowed us to combine budget and spiraling approaches to evaluate the role of hydrologic variability as it controls nutrient cycling. Our approach was to calculate a budget of ^{15}N by measuring standing stocks and export of ^{15}N tracer using a mass-balance approach (Tank et al. 2000), and by measuring export as particles at the end of the reach through 1 yr following each isotope addition. We also related N cycling parameters to changes in stream water and groundwater exchange during the two seasons.

Methods

Study stream—Spring Creek (44.29°N, 115.25°W) is a second-order mountain stream in the Payette River drainage, at an elevation of 2116 m in the Sawtooth Mountains of Idaho, U.S.A. (Fig. 1). The 1.9-km study reach starts in a sparse forest dominated by lodgepole pine (*Pinus contorta*) that gradually grades into a riparian meadow and delta plain dominated by willows (*Salix* spp.) and sedges (*Carex* spp.; Arp et al. 2006; Fig. 1) and ends at Bull Trout Lake. This reach occurs in a glacial outwash valley with the stream slope gradually declining from 0.007 m m^{-1} to 0.003 m m^{-1}. The reach is normally covered with snow from December through April. The stream has a typical snowmelt-driven hydrograph (Fig. 2), with peak flows near 01 June of each study year. Mean wetted width was 3.8 m during snowmelt flows and 3.1 m during baseflow. Substrate is composed of pebbles, gravels and sand, with a median size (D_{50}) of 11 mm (Arp et al. 2007). The spring-fed creek was cold, with mean temperatures during our June and August tracer additions of 4.6°C and 7.3°C, respectively. Nitrate concentrations were low, 10.7 μg N L^{-1} during the snowmelt addition and 9.3 μg N L^{-1} during baseflow. Phosphate phosphorus concentrations were near detection limit (1 μg P L^{-1}).

Fig. 1. Spring Creek near where it exits forest onto a glacial outwash floodplain. Note the dry gravel bar where we sampled parafluvial sediment.

Hydrology—Stream depth was measured hourly using pressure transducers at gauging stations located at the addition site (0 m) and 1559 m downstream in 2002 and 2003, with additional stations at 900 and 1865 m downstream in 2004. At each station, we developed a stage–discharge relationship (Arp et al. 2006).

To measure the degree of exchange between stream water and groundwater, we installed sampling wells in the channel and riparian zone. These wells were augered to 1.5-m depth and cased with 5-cm-diameter machine-slotted (0.25 mm) polyvinyl chloride (PVC) well screen below the water table and finished to the top with solid PVC. Wells were backfilled with native material, capped with a bentonite clay plug, and purged before sampling. In-channel wells were installed to 50-cm depth into gravel alluvium with a rod and cased with 2.5-cm-diameter PVC pipe. Sampling transects, consisting of stream water, an in-channel well, and two lateral wells (4–30 m from stream), were located at 150, 300, 800, and 1500 m downstream of the addition site. Groundwater levels in the wells were measured using an electronic beeper attached to a ruler. To estimate mixing of stream water in wells, we sampled for Br^- in all wells before each experiment and on day 14. Stream water was labeled with Br^- from a continuous injection (*see* below). Hyporheic mixing was calculated as the ratio of groundwater to stream water Br^-, less background concentration (Triska et al. 1989). We constructed subsurface flow nets (Freeze and Cherry 1979) by measuring water-surface elevations of the stream

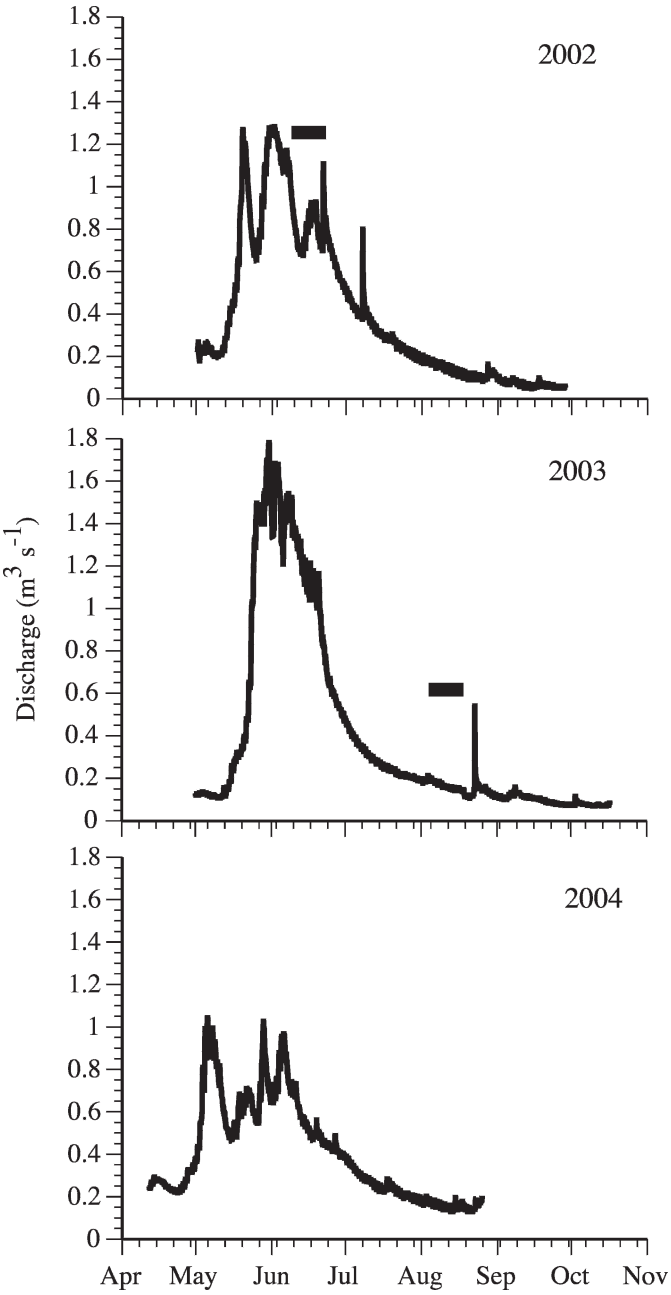

Fig. 2. Hydrographs of Spring Creek during the 3 yr of this study. Bars indicate the period of the 2-week snowmelt and baseflow ^{15}N additions.

surface, backwaters, and isolated ponds using a total station laser theodolite.

^{15}N additions—We added ^{15}N as 28 atom-% $Na^{15}NO_3$ for 2 weeks during snowmelt from 13 June to 27 June 2002 and during baseflow from 05 August to 19 August 2003 1895 m upstream from the lake (Fig. 2). For the snowmelt addition we added 70 g ^{15}N, and 56 kg of Br^- (as NaBr) as a conservative tracer of water. For the baseflow addition we added 75 g ^{15}N and 12.8 kg of Br^-. Calculated enrichments of δ^{15} NO_3^- were 1704‰ during snowmelt and 11,670‰ during baseflow. Solutes were mixed daily into a 20-liter carboy and continuously pumped into the stream at 10.4 mL min^{-1} using a Watson–Marlow peristaltic pump (Watson–Marlow). To measure removal, storage, and residence times of N in the reach we collected samples periodically at 62, 120, 225, 539, 1069, 1560, and 1895 m below the addition site.

Solute sampling—We measured the removal of ^{15}N-NO_3^- from the water column on days 3, 7, and 14 following the start of each experiment. We collected 10 liters of water for analyses of ^{15}N-NO_3^-, ^{15}N-ammonium (^{15}N-NH_4^+), and total dissolved ^{15}N (^{15}N-TDN) at each site. To estimate NO_3^- regeneration and/or transformation to NH_4^+ and dissolved organic N (DON) we measured dissolved ^{15}N pools at each station on the day after the injection, then weekly or biweekly thereafter at the top and bottom of the stream reach. For ^{15}N-NO_3^-, 3-liter samples were spiked with 150 μg NO_3-N and concentrated to 0.1 liter by boiling. Devarda's alloy catalyzed conversion of NO_3^- to NH_4^+ during a 48-h incubation (Mulholland et al. 2008). ^{15}N-NH_4^+ was collected on an acidified filter and $\delta^{15}N$ and N mass were measured at the University of California Davis Stable Isotope Facility. We measured ^{15}N-NH_4^+ in 3-liter unamended samples using ammonia diffusion (Peterson et al. 2001). We measured ^{15}N-TDN by persulfate oxidation of 0.75-liter samples, followed by conversion of NO_3^- to NH_4^+ following the same procedure for ^{15}N-NO_3^- above, and calculated ^{15}N-DON as the difference between ^{15}N-TDN and ^{15}N-NO_3^- + ^{15}N-NH_4^+.

We measured stream water NO_3^- and Br^- concentrations on a DIONEX-500 ion chromatograph with concentrator and AS14 analytical and guard columns. Detection limits were 0.5 μg L^{-1} NO_3-N and 2 μg L^{-1} Br^-. Water samples for TDN concentration were oxidized using persulfate digestion (Valderrama 1981) followed by NO_3-N quantification on the digested samples using second-derivative spectroscopy (Crumpton et al. 1992), with a detection limit of 15 μg L^{-1}. NH_4-N concentration was measured fluorometrically (Holmes et al. 1999) with a detection limit of 2 μg L^{-1}.

Particle sampling—We collected samples for both ^{15}N and standing stock of N during and after each addition to sequentially inventory ^{15}N in the benthos. At each of the seven sites we sampled the major benthic stocks of N. All particle samples were analyzed at the University of Wyoming Light Stable Isotope Facility using continuous-flow mass spectrometry. Samples were combusted in an elemental analyzer (from which we could calculate mass of N and C), linked to a VG IsoPrime or Finnegan Delta Plus mass spectrometer.

Fine benthic organic N (FBON) was sampled with a 20.3-cm-diameter stovepipe corer by setting the corer into sediment, swirling the sediment, and subsampling the suspended water for ^{15}N and organic matter as ash-free dry mass (AFDM). We filtered subsamples through Gelman A/E filters for both ^{15}N and organic matter quantification. In 2002 we separated surface from deep (to 10 cm) FBON. There was no difference in the $\delta^{15}N$ of

surface and deep FBON in 2002; therefore, in 2003 and 2004 we collected only one sample that combined surface and deep FBON. We measured organic matter standing stock (as AFDM) and calculated N standing stock using the measured C : N ratio and assuming that organic matter is 50% C. The ^{15}N in the samples was extrapolated to a ^{15}N standing stock (g ^{15}N m^{-2}) by multiplying atomic ratio of ^{15}N excess ($^{15}N_{xs}$, which is background-corrected atomic ratio of ^{15}N) of FBON by the standing stock of N at each site. We multiplied standing stock by reach-wetted area to extrapolate to the reach.

We sampled N from rock biofilms (epilithon) by collecting all stones in a 20.3-cm core and scrubbing epilithon into a slurry using a detail brush. Processing and scaling of AFDM and ^{15}N from this slurry was as for FBON samples, as was scaling ^{15}N standing stock. We also measured chlorophyll *a* (Chl *a*) by filtering 1–5 mL of slurry onto Gelman A/E filters, extracting in 90% basic ethanol, and measuring on a calibrated fluorometer.

We sampled invertebrates at each site by collecting and freezing several individuals of four dominant taxa for ^{15}N analysis. Taxa were *Yoraperla*, a shredder stonefly; *Sweltsa*, a predatory stonefly; *Drunella* and *Cinygmula*, scraper mayflies. These four taxa constituted 44% to 67% of total assemblage biomass depending on year. For each isotope addition in 2002 and 2003, and in July 2004, we measured biomass of all taxa from two composite 20.3-cm-diameter stovepipe core samples at each of seven sites, following Hall et al. (2006). From these stovepipe core samples we also measured standing stock of coarse benthic organic matter (CBOM) as AFDM, which was mostly terrestrial leaf litter. We calculated $^{15}N_{xs}$ for CBOM as for FBON and scaled the value to the stream reach.

After the 2002 snowmelt addition we discovered that hyporheic storage of N was likely to be a large sink for N, so we sampled the shallow parafluvial zone to estimate hyporheic storage of N in this region. We sampled 18 gravel bars adjacent to the stream six times following the 2003 baseflow addition (Fig. 1). We dug a hole to the water table in a dry bar, and we inserted a plastic, 8.2-cm-diameter corer 9–20 cm deep into the sediment beneath the water surface. We plugged the top, excavated the core, and dropped the contents into a bucket with a measured amount of stream water. We stirred the sample to suspend particulate organic matter and sampled particulate organic matter as AFDM and ^{15}N from the resultant slurry. We extrapolated ^{15}N standing stocks by multiplying area-specific mass of $^{15}N_{xs}$ by the bankfull area measured in 2002 as part of a wood survey.

We measured volume and surface area of wood in the stream by using the line-intercept technique (Wallace and Benke 1984) once during each ^{15}N addition. We established a transect every 25 m through the 1.9-km study reach and measured the diameter of each piece of submerged wood. To sample for ^{15}N, we sampled a measured area of loosely attached wood biofilm from each of three sticks. We dried and weighed the sample and subsampled for ^{15}N and %N measurements. We calculated the mass of $^{15}N_{xs}$ per area of wood at each site by multiplying $^{15}N_{xs}$ by mass of N per unit area of wood biofilm. We scaled this value to the entire reach by multiplying by the total surface area of wood.

During the wood survey, we measured the percent cover of the limited macrophyte vegetation and standing stocks of five areas where macrophytes constituted 100% cover to estimate a reach-scale mass of macrophytes. On day 14 we sampled macrophytes at the seven sample sites for ^{15}N and %N.

We measured enrichment of riparian willows at the end of the ^{15}N addition in August 2003. We collected leaves from three locations immediately adjacent to the stream upstream from the addition site and 14 locations downstream and measured their ^{15}N.

We measured suspended particulate N (seston) flux at the bottom of the reach (1895 m) ~ weekly following the ^{15}N additions during June–October of each year. During Spring 2004 we used an autosampler located at Sta. 6, (1560 m) to capture samples during the snowmelt flood. For each sample we filtered a known volume of water onto 25-mm Gelman A/E filters, dried them, and analyzed them for both total N and ^{15}N. Instantaneous seston export was calculated as the $^{15}N_{xs}$ of seston times discharge times concentration of seston N.

We fortuitously sampled seston export during one storm in each summer: one during the snowmelt addition and the other 2 d following the baseflow addition (Fig. 2). In the snowmelt addition we hand-collected eight samples during and following a storm that increased discharge from 0.7 m^3 s^{-1} to 1.1 m^3 s^{-1}. Just following the baseflow addition we installed an ISCO auto sampler at the bottom of the reach that sampled every 2 h, 19 times during a storm that increased discharge from 0.11 m^3 s^{-1} to 0.55 m^3 s^{-1}.

Denitrification—In 2002 we measured potential denitrification rates in situ using a modified acetylene block method (Baker and Vervier 2004) at three surface parafluvial sites (gravel bars or abandoned side channels) and two subsurface parafluvial sites (wells). For surface sites, we excavated sediments and inserted a 20.3-cm-diameter stovepipe 20 cm below the water level. Water from the stovepipe and wells was pumped to a carboy where it was bubbled with acetylene gas and mixed with Br^-, glucose, and NO_3^- to final concentrations of 0.07 mg Br^- L^{-1}, 1.0 mg C L^{-1}, and 0.25 mg N L^{-1}. We pumped this water back into the pipe or well and collected samples for N_2O gas and Br^- via a syringe attached to the peristaltic pump. Samples for N_2O gas were stored in evacuated glass vials for later analysis by gas chromatography. Br^- concentrations were used to correct N_2O concentrations for dilution and denitrification potential was calculated as the slope of the regression line of dilution-corrected N_2O concentration vs. time (Baker and Vervier 2004).

During both snowmelt and baseflow additions we collected stream water for analysis of $^{15}N_2$ and $^{15}N_2O$ as products of denitrification (Mulholland et al. 2008, 2009). In 2002 only two samples were analyzed from samples collected at the top and bottom of the study reach on day 12 of the tracer addition. In 2003, samples were collected in duplicate from each station on days 2, 4, and 14. In all

cases, samples were collected via a 60-mL syringe, and were stored under water until headspace equilibration with ultrapure helium following Mulholland et al. (2008). Gas samples were analyzed for ^{15}N by mass spectrometry at the University of California Davis Stable Isotope Facility.

Data analysis—For display purposes we show $\delta^{15}N$, but for all calculations we used the atomic ratio excess ($^{15}N_{xs}$) of ^{15}N, which corresponds to the ratio of masses, $^{15}N/(^{14}N + {}^{15}N)$, from which the unlabeled background atomic ratio is subtracted. We measured background ^{15}N prior the 2002 addition and from samples upstream of the addition site.

We calculated flux of ^{15}N as NO_3^- (mg N min^{-1}), $F_{15NO_3\,x}$ at each station (x) as

$$F_{15NO_3\,x} = {}^{15}N_{xs\,NO_3} \times [NO_3]_x \times Q_x \quad (1)$$

where Q_x is the discharge at that station as calculated by mass balance of the added Br^- conservative tracer. Per-meter removal (same as uptake) rate (k) is calculated by fitting ^{15}N flux at each station to the following model using least-squares regression:

$$\ln F_{15NO_3\,x} = \ln F_{15NO_3\,0} - kx \quad (2)$$

The inverse of k is the uptake length of nitrate (S_{NO_3}), which is the average distance a NO_3^- ion travels before removal from the water column. Stream depth and velocity in part, control uptake length (Hall et al. 2009), so we calculated uptake velocity (v_f, m min^{-1}) to compare nutrient uptake at high and low discharge:

$$v_f = Q/(w\, S_{NO_3}) \quad (3)$$

where w is wetted channel width. We calculate uptake flux of NO_3^- (U_{NO_3} mg N m^{-2} min^{-1}) as

$$U_{NO_3} = v_f \times [NO_3] \quad (4)$$

To estimate a mean and confidence interval for uptake parameters for the two baseflow dates, we pooled data from both sampling dates because slopes and intercepts of the regression (Eq. 2) did not differ (ANCOVA with interaction term) and we calculated a confidence interval of the slope from the pooled regression. Slopes were significantly different in the snowmelt addition so we simply calculated a 90%, two-tailed, t-based confidence interval with $n = 3$ measurement dates. To estimate uncertainty in mass of ^{15}N stored in the reach, we calculated the 90% confidence interval of the mean of the $^{15}N_{xs}$ mass ($n = 7$ for most compartments, $n = 14$–17 for hyporheic N). We calculated the propagated uncertainty for the sum of all compartments assuming no covariance among the individual compartments. If ^{15}N stocks declined with time we calculated residence time (d) as the inverse slope of ln ^{15}N mass ($g^{15}N_{xs}$ $reach^{-1}$) vs. time.

Results

Hydrology—Spring Creek had a snowmelt-driven hydrograph during the three study years (Fig. 2). During the snowmelt addition, discharge declined from 0.75 m^3 s^{-1} to 0.61 m^3 s^{-1}. A rain-on-snow storm on 21 June 2002 increased discharge to 1.10 m^3 s^{-1} (Fig. 2). During the baseflow addition, stream discharge declined evenly from 0.17 m^3 s^{-1} to 0.12 m^3 s^{-1} (Fig. 2).

Water-table maps for the lower 500 m of the stream reach during snowmelt and baseflow showed seasonal differences in surface water–groundwater exchange. During snowmelt the stream-surface elevation exceeded that of the riparian zone; thus, flow direction was from the stream to the floodplain and hyporheic zone (Fig. 3A). During baseflow, stream elevation was similar to the near-stream water table, with much of the subsurface flow parallel to the direction of stream flow, but with strong and isolated areas of water flow to the hyporheic zone (Fig. 3B). These results were corroborated by observation of Br^- in sampling wells (Fig. 4). Overall, the fractions of hyporheic water that were derived from stream water were highest during baseflow (10–70%) compared to snowmelt (1–40%) and were higher in in-channel wells than lateral wells in all but one site (Fig. 4).

Uptake and transformation—Of the 70 g ^{15}N added to Spring Creek during snowmelt, 24.5 g, or 35%, was removed from stream water before the end of the 1.9-km reach. The balance, 45.5 g ^{15}N, entered the lake at the end of the stream reach, and is no longer considered in any calculations. During baseflow, 43.5 g ^{15}N or 58% of 75 g added was removed from the stream water (Table 1). Uptake lengths were two times longer during snowmelt than during baseflow (Table 1). Uptake velocity was about 70% higher during snowmelt, although the 90% confidence intervals overlapped (Table 1).

We were able to collect enough water for ^{15}N analysis from only one well during each addition. During snowmelt, the well contained 16% stream water, yet only 3% of the ^{15}N concentration relative to the nearest stream sampling location, indicating 82% removal. During baseflow, the percent stream water was much higher (97%) and 51% of the stream water ^{15}N concentration was detected indicating 50% removal.

^{15}N in epilithon was unrelated to sampling location during the snowmelt addition, in part because long uptake lengths meant that decline in tracer concentration with distance was small. However, variation in epilithon $\delta^{15}N$ was strongly positively correlated with Chl *a* standing stock suggesting that biofilms with higher photosynthetic biomass had higher N uptake (Fig. 5). During the baseflow addition, a downstream decline in $\delta^{15}N$ of epilithon was explained by distance from the addition site. The discharge-corrected rate of decline was 0.00061 m^{-1}, close to the measured uptake rate of ^{15}N-NO_3^- on day 14 (Table 1) showing that enrichment of benthic pools mirrored the removal rate we measured by using water column nitrate.

Little of the added ^{15}N-NO_3^- was transformed to other dissolved pools. One day following both additions there was detectable label in NO_3^- which, when scaled for 15 d, corresponded to a small amount of exported ^{15}N (Table 2). During snowmelt, NH_4^+ and DON pools were not enriched by day 14 of the ^{15}N addition. In contrast, during baseflow,

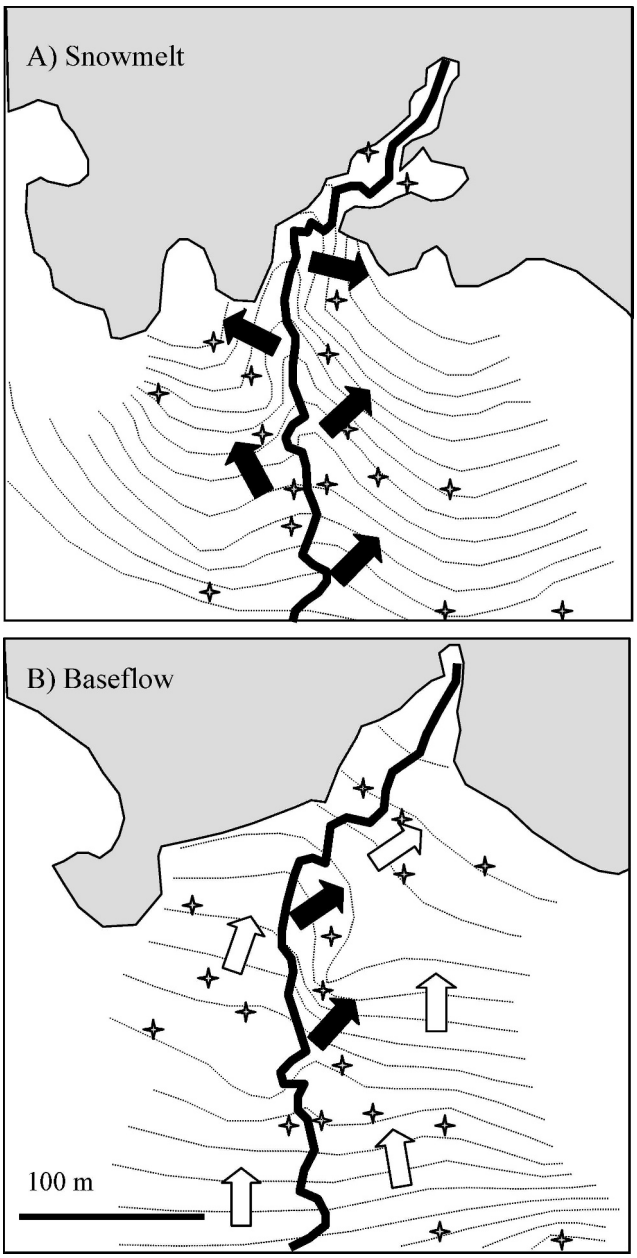

Fig. 3. Water-table maps showing higher flow of stream-water to the subsurface zone during (A) snowmelt compared to (B) baseflow. Predominance of black arrows during snowmelt show the stream loses water to the hyporheic zone, while during baseflow, white arrows show that most of the subsurface flow is parallel to the stream. Stars indicate water-table monitoring points. Dotted lines represent 0.1-m contours of water-table height above an arbitrary datum descending to Bull Trout Lake.

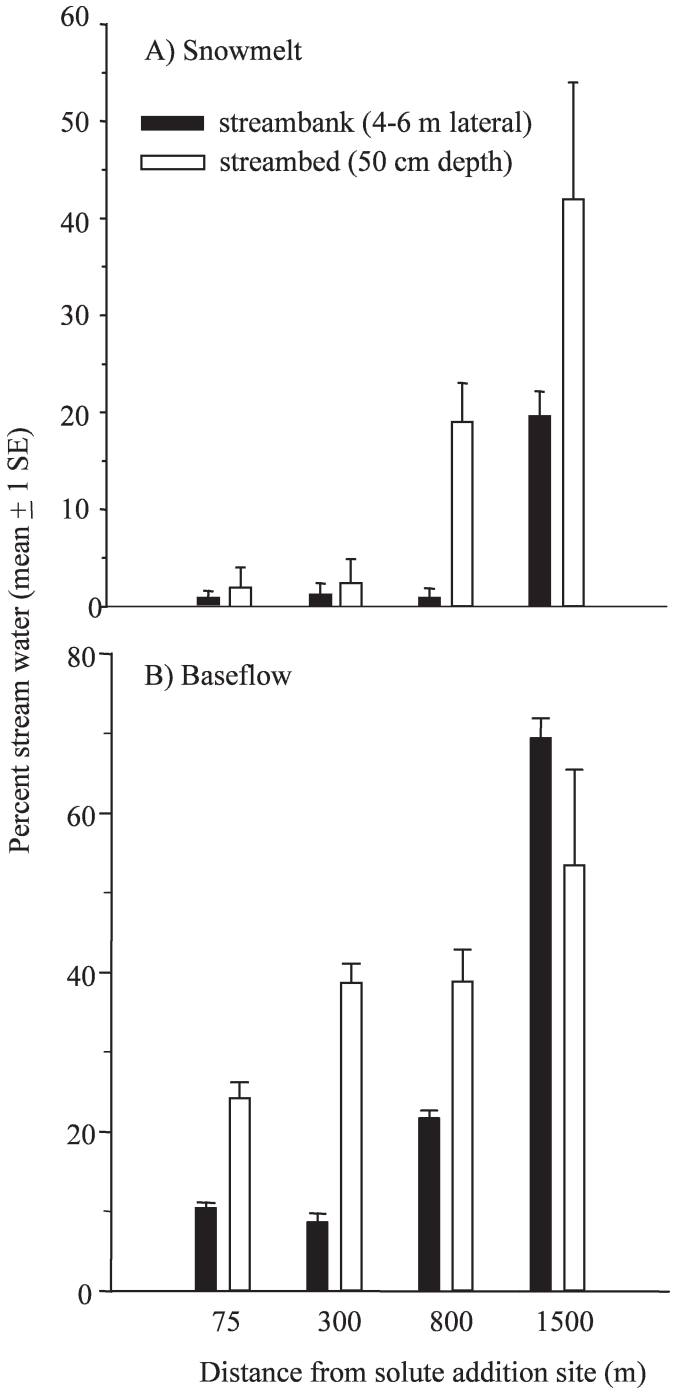

Fig. 4. Percent stream water measured from Br^- in wells located in the channel and lateral to the stream show substantial hyporheic exchange during both experiments.

the NH_4^+ pool was enriched by 147‰ on day 14, which represents 0.0026 μg ^{15}N L^{-1}. If NH_4^+ were this enriched for the 2-week period, then ^{15}N-NH_4^+ export would have been 0.5 g. Similar to the snowmelt addition, the DON pool was not enriched during the baseflow addition. We can constrain the amount or tracer N exported as DON in the baseflow addition. If DON contained the minimal observable label of 4‰ above background, and given DON concentration of 51 μg N L^{-1} during the 2-week addition, DO^{15}N could have contributed 0.14 g ^{15}N of export, which is smaller than ^{15}N-NH_4^+ export, and 20-fold smaller than seston export (Table 2).

Denitrification accounted for little N loss during both tracer additions. During snowmelt, denitrification potential was not detectable in parafluvial sediments, and potential rates were low in the two wells (mean and SE = 0.052 ±

Table 1. Nitrate uptake during the snowmelt and baseflow isotope additions. Fraction removed describes the amount of ^{15}N removed by the 1895-m stream reach relative to what we added at the measurement time; the balance was exported to the lake. No data are shown for baseflow at the 3-d sampling period because regression of dilution-corrected ^{15}N flux vs. distance downstream was not statistically significant; thus, uptake rate could not be calculated. Ninety percent confidence intervals are shown below each value in parentheses.

^{15}N addition	Day	Discharge ($m^3\ s^{-1}$)	Uptake rate (1000 m^{-1})	Uptake length (m)	Uptake velocity (mm min^{-1})	Fraction removed
Snowmelt	3	0.754	0.134(0.069–0.198)	7460(5040–14300)	1.60	0.22
	8	0.576	0.312(0.164–0.461)	3210(2170–6100)	2.84	0.45
	14	0.488	0.248(0.207–0.289)	4030(3460–4810)	1.91	0.37
	$\bar{x}$		0.232(0.045–0.28)	4320(740–5640)	2.12(1.24–3.36)	0.35(0.19–0.53)
Baseflow	8	0.160	0.369(0.081–0.658)	2710(1520–12410)	1.15	0.50
	14	0.127	0.528(0.408–0.648)	1894(1540–2450)	1.30	0.63
	$\bar{x}$		0.456(0.338–0.573)	2190(1740–2950)	1.27(0.95–1.60)	0.58(0.47–0.66)

0.012 μg N_2O-N L^{-1} min^{-1}). Dissolved gas pools were not labeled; mean $\delta^{15}N_2$ and $\delta^{15}N_2O$ collected during tracer test plateau were not different from background samples (*t*-test, N_2 $t = 0.75$, df = 37, $p = 0.46$; N_2O $t = 0.49$, df = 36, $p = 0.63$). Given that $\delta^{15}N\text{-}NO_3^-$ in the baseflow addition was about 60% of enrichments in studies designed to measure denitrification (Mulholland et al. 2009), this level of enrichment would have reduced our ability to detect enriched N_2 by < one-half.

Storage—Both snowmelt and baseflow additions strongly enriched benthic pools. During the snowmelt addition FBOM was enriched by 13‰, eplilithon 220‰, wood 240‰, and invertebrates 250‰. Baseflow addition enrichments were 5–10 times higher.

Fine benthic organic N constituted a substantial stock of both total N and tracer ^{15}N in the stream (Table 2). FBON standing stock and tracer ^{15}N were higher during baseflow than snowmelt. The standing stock of FBON $^{15}N_{xs}$ stayed roughly constant for the 60–80 d following the snowmelt addition (Fig. 6). For the baseflow addition, FBON $^{15}N_{xs}$ remained nearly constant through the autumn and winter (Fig. 6). However, in both additions the ^{15}N standing stock of this pool declined greatly following the subsequent snowmelt flood with zero $^{15}N_{xs}$ remaining by August 2003, 1.2 yr following the snowmelt addition and 0.6 g $^{15}N_{xs}$ in August 2004, 1 yr following the baseflow addition. Because storage was constant until the following snowmelt, we did not calculate a residence time for FBON.

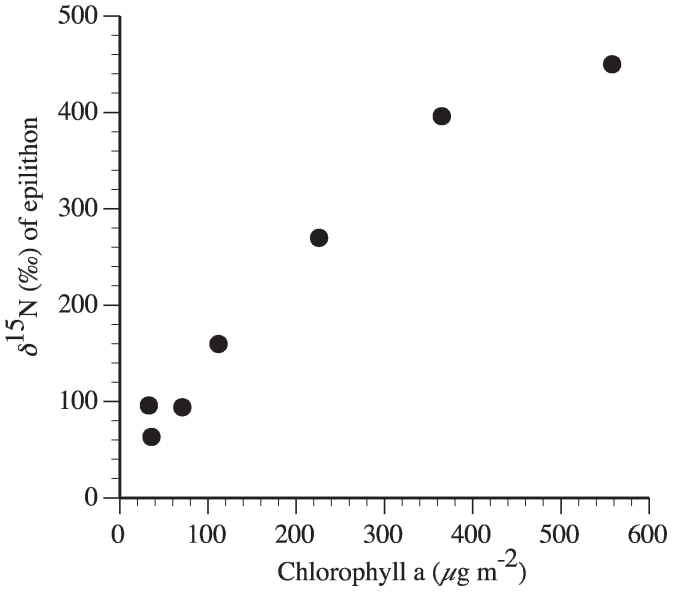

Fig. 5. During the 2002 snowmelt addition, epilithon $\delta^{15}N$ increased as a function of epilithon chlorophyll standing stock. Data are from day 14, which was the end of the snowmelt ^{15}N addition.

Epilithon had low standing stock of N, but high label (average $\delta^{15}N$ on day 14 in the snowmelt and baseflow additions were 218‰ and 1610‰, respectively) such that it represented a stock of $^{15}N_{xs}$ about equal to the much larger FBON pool (Table 2). Mass of epilithic tracer ^{15}N declined with time immediately following the baseflow addition, but not the snowmelt addition (Fig. 6). Tracer ^{15}N stock in epilithon was zero in summer 2003, a year after the snowmelt addition, and only 0.2 g the summer following the baseflow addition, showing that epilithon was not a substantial long-term store of N. Net residence time of epilithon $^{15}N_{xs}$ was calculated for only the baseflow addition and was 117 d.

Wood volume was similar between 2002 and 2003, with 0.0032 $m^3\ m^{-2}$ and 0.0037 $m^3\ m^{-2}$ of channel area, respectively. Surface area of wood was 0.095 in 2002 and 0.125 $m^2\ m^{-2}$ in 2003. Because the N content of wood biofilm was low, standing stock of N was low relative to other pools (Table 2). Wood biofilm contained a small amount of tracer N mass at day 14 (Table 2), and this $^{15}N_{xs}$ declined exponentially with time following both additions, with an estimated net residence time of 166 d following snowmelt addition and 117 d (coincidentally the same as for epilithon) following the baseflow addition.

Hyporheic standing stocks of N measured in the parafluvial zone in 2003 were much higher than all other pools combined (Table 2). Isotopic enrichment was considerably lower than other pools averaging 31‰ during the three sampling dates following the baseflow addition in 2003. However, because standing stock was so high, parafluvial organic matter contained a large amount of tracer N following the addition. Standing stock of ^{15}N in summer 2004 was not different than that following the ^{15}N

Table 2. Mass of ^{15}N excess ($^{15}N_{xs}$) in benthic pools and seston for the two additions at the end of each tracer injection (day 14). Hyporheic fine benthic organic N (FBON) is the average of three dates collected in summer and autumn 2003. 'Percent of removed accounted for' is the total $^{15}N_{xs}$ inventoried in the reach relative to the amount of $^{15}N\text{-}NO_3^-$ removed by the stream during the 14 d addition (i.e., not including what was exported to the lake as unremoved $^{15}N\text{-}NO_3^-$). NM means not measured.

	Snowmelt		Baseflow	
N pool	N stock (mg N m^{-2})	Tracer stock (g ^{15}N $reach^{-1}$)	N stock (mg N m^{-2})	Tracer stock (g ^{15}N $reach^{-1}$)
Fine benthic organic N	200	0.1	1200	4.3
Epilithon	40	0.3	110	4.0
Wood biofilm	17	0.1	14	0.3
Coarse benthic organic N	110	0.3	120	0.4
Macrophytes	NM	NM	20	0.5
Insects	72	0.3	82	1.3
Hyporheic FBON	NM	NM	6100	4.1
Seston export to day 14		1.5		2.6
NH_4^+ export to day 15		0*		0.5
NO_3^- export to day 15†		0.5		0.1
DON export to day 15		0*		0*
Denitrification		0*		0*
Total		3.1		18.1
90% CI		(2.3–3.9)		(11.8–23.7)
% of removed accounted for		13		42

* ^{15}N enrichment not detectable above background.

† Export only includes NO_3^- that was mineralized and not tracer that was directly exported from the reach during the 14-d addition without being taken up.

addition in 2003, showing that this store of ^{15}N was not removed by the snowmelt flood (Fig. 7). We cannot calculate residence time using an exponential model for this hyporheic N because the standing stock of $^{15}N_{xs}$ did not decline with time, but residence time was much $>$ 1 yr.

Willows collected from near the stream had a variable pattern of labeling from 42‰ to background (Fig. 8). Because of the spatial patchiness of labeling and not knowing the mass of potentially labeled willows, we could not calculate the tracer ^{15}N mass stored in willows; however, enriched willow leaves show that hyporheic flow transported stream water $^{15}N\text{-}NO_3^-$ into riparian pools, which was used by vegetation.

CBOM and macrophytes constituted only a small fraction of inventoried $^{15}N_{xs}$ immediately following both additions (Table 2).

Insects constituted a substantial stock of N, despite having relatively low biomass (0.7–1 g AFDM m^{-2}; Table 2), but did not constitute a long-term store of N. By the following summers, insects contained 0 (snowmelt addition) or 0.04 g $^{15}N_{xs}$ (baseflow addition).

Seston export—Seston ^{15}N export during the snowmelt addition was highest at the end of the 14-d addition and declined rapidly afterward because of decreasing $\delta^{15}N$ of the seston coupled with falling stream discharge (Figs. 2, 9). The storm during the snowmelt addition exported 0.3 g $^{15}N_{xs}$. Although seston was still labeled during late summer and autumn, export was low. Total $^{15}N_{xs}$ export as seston was 2.7 g (including the storm), which was 11% of the 24.5 g $^{15}N_{xs}$ removed from the water column in the reach. We did not measure seston export during the snowmelt flood the year following the addition. Label at day 80 was 12‰ above background and undetectable in one sample during the 2003 snowmelt flood. If seston were enriched by 6‰ during snowmelt 2003, then based on discharge and estimated particle N concentration, export could have been 2 g ^{15}N. Cumulative export of seston particles through 2003 was about 2.5 times higher than the total amount measured in benthic pools on day 14 of the snowmelt addition showing that these particles were coming from an unmeasured pool.

As a proportion of ^{15}N removed, Spring Creek exported 6.5 times more ^{15}N as seston in the baseflow addition relative to the snowmelt addition (Fig. 9; Table 3). Like the snowmelt addition, export was highest immediately following the baseflow addition and declined through autumn reflecting declines in both seston $\delta^{15}N$ and discharge (Figs. 2, 9). Seston $\delta^{15}N$ and discharge declined throughout autumn (Figs. 2, 9). During late summer and autumn the stream exported 12.1 g $^{15}N_{xs}$ of seston, which included the storm on day 18. Export presumably was low during the winter because of low discharge. Given $\delta^{15}N$ of seston in autumn and a constant discharge of 0.07 m^3 s^{-1} throughout the winter, we estimate winter seston flux as no higher than 2.4 g $^{15}N_{xs}$. At the onset of snowmelt in spring 2004, discharge increased, driving renewed export of particulate ^{15}N. Due to higher flow, seston export during the following snowmelt was 16.1 g $^{15}N_{xs}$, which was higher than export during the previous summer and autumn. Total seston export following the baseflow addition was 31.0 g, which represented 71% of the $^{15}N_{xs}$ removed from the stream during the addition (Table 3).

The storm on day 18 during the baseflow addition had high instantaneous rates of seston export, but it represented only 8% of total labeled seston export from the reach

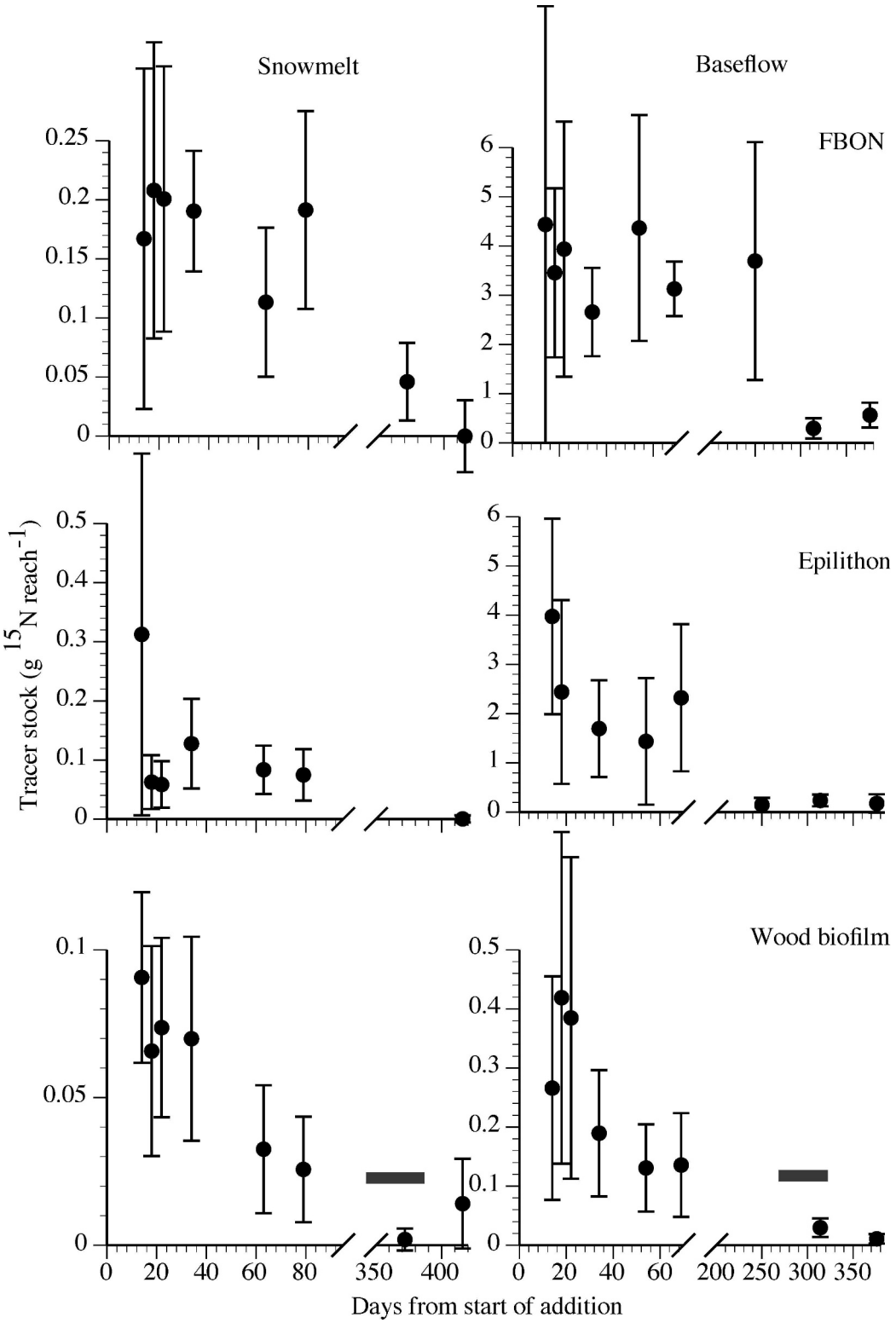

Fig. 6. Mass of $^{15}N_{xs}$ in FBON, epilithon, and wood biofilms during the snowmelt and baseflow additions. Note differences in Y-axis scaling, indicating the much higher tracer mass during the baseflow addition than during the snowmelt experiment. The tracer also persisted in the baseflow addition relative to the snowmelt addition. Bars on each figure indicate the snowmelt flood following the additions the previous year. Y-axis is g $^{15}N_{xs}$ in a particular pool scaled to the entire reach. Error bars are 90% *t*-based confidence intervals of the mean.

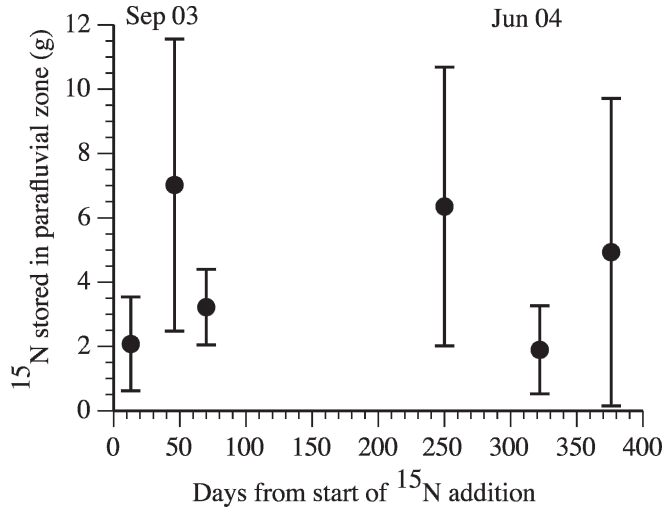

Fig. 7. Hyporheic storage of $^{15}N_{xs}$ did not decline with time for 1 yr following the baseflow ^{15}N addition in 2003. Y-axis is g $^{15}N_{xs}$ in the entire stream reach of hyporheic zone immediately beneath and adjacent to the wetted area of the stream, but within the active channel. Error bars are 90% *t*-based confidence intervals of the mean.

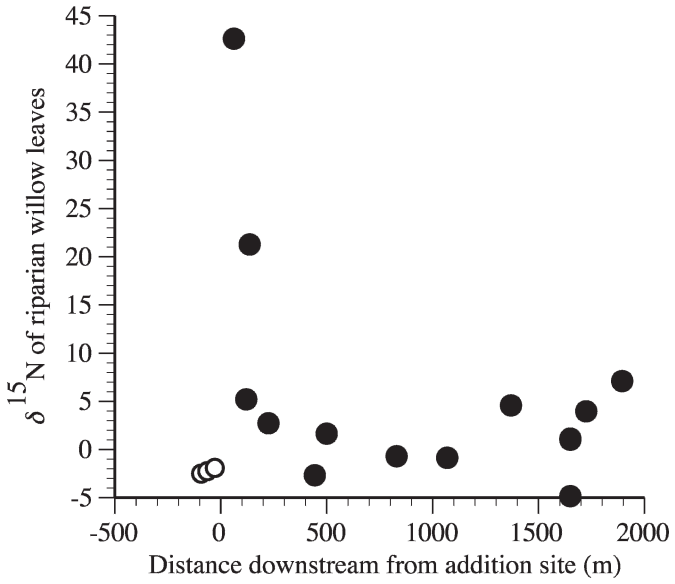

Fig. 8. Some willows downstream of the addition site contained ^{15}N label. X-axis is the distance downstream of the addition site, and Y-axis is the $\delta^{15}N$ of willow leaves. Open symbols are from three unlabeled willows upstream of the addition site. Solid symbols are willows that could potentially be labeled.

(Figs. 9, 10). Particle N concentrations increased 1000-fold during this storm (Fig. 10B), and the seston $\delta^{15}N$ declined to about 2‰ above background (Fig. 10C), suggesting that the high N export was not of labeled, benthic N, but rather particle N from upstream or riparian areas. As particle N concentration dropped, $\delta^{15}N$ rebounded, and even exceeded prestorm values as exported N reverted to N from the channel bottom in the study reach. Total export of tracer $^{15}N_{xs}$ as seston during this storm (Fig. 10D) was 2.4 g $^{15}N_{xs}$, which represented 16% of the 15 g of $^{15}N_{xs}$ inventoried on the stream bed 3 d earlier (Table 2).

Mass balance—Of the 70–75 g ^{15}N-NO_3^- added each season, 24.5 g was removed from the water column during snowmelt season and 43.5 g was removed during baseflow season. The remainder was exported as NO_3^- to Bull Trout Lake. Of the ^{15}N-NO_3^- that was removed from the water column, the fraction found in benthic pools was much lower in the snowmelt compared to the baseflow addition (Table 3). By day 14 of the snowmelt addition we recovered only 13% of the $^{15}N_{xs}$ that was removed from the water column, but much more (42%) in the baseflow addition (Table 2). We did not sample the parafluvial zone in the snowmelt addition, but assuming that the fraction of N transported into this zone was similar to that in the baseflow addition, we would not have increased the inventory by much.

Total seston export following each addition accounted for 11% of the N removed during snowmelt and 71% of N during baseflow (Table 3). Much (39%) of ^{15}N removed during the baseflow addition exited the stream during the next snowmelt flood. For both additions, the total amount of N exported as seston exceeded the amount inventoried at any one time in the benthic pools, showing that an unaccounted pool must have supplied seston for export.

Discussion

Spring Creek was highly retentive of N, both during snowmelt and baseflow hydrologic regimes, with long residence times of benthic pools and unmeasurable (i.e., > 1 yr) turnover of parafluvial sediment pools of N. This high retention was despite a large annual snowmelt flood and streambed composed of gravels with high mobility during such flows (Arp et al. 2007; Myers et al. 2007). Thus, N residence time in the stream is on the order of ≥1 yr, possibly longer, and strongly points to storage as an important fate for N that has been assimilated from the water column.

Benthic uptake, hydrologic storage, and denitrification—Nitrate removal, measured as uptake velocity was similar between the snowmelt addition and the baseflow addition. Demand for this N was high; v_f value during snowmelt was in the top 20% of 72 tracer experiments in streams (Hall et al. 2009). High assimilatory N removal in Spring Creek is perhaps not surprising given its low dissolved inorganic N concentrations (5–20 μg N L^{-1}) and high demand for this nutrient. However, high demand for N during snowmelt is somewhat surprising because streams have higher biofilm biomass during baseflow than during or after floods (Fisher et al. 1982), and this higher biomass should take up NO_3^- more rapidly. During the baseflow experiment epilithon and FBON standing stocks were higher and temperatures were warmer than during snowmelt. In fact, benthic uptake of N was higher in the baseflow addition (after correcting for higher ^{15}N in the water) and, thus, we might have expected uptake velocity to be higher because of this higher biological demand. However, the similar uptake velocity in

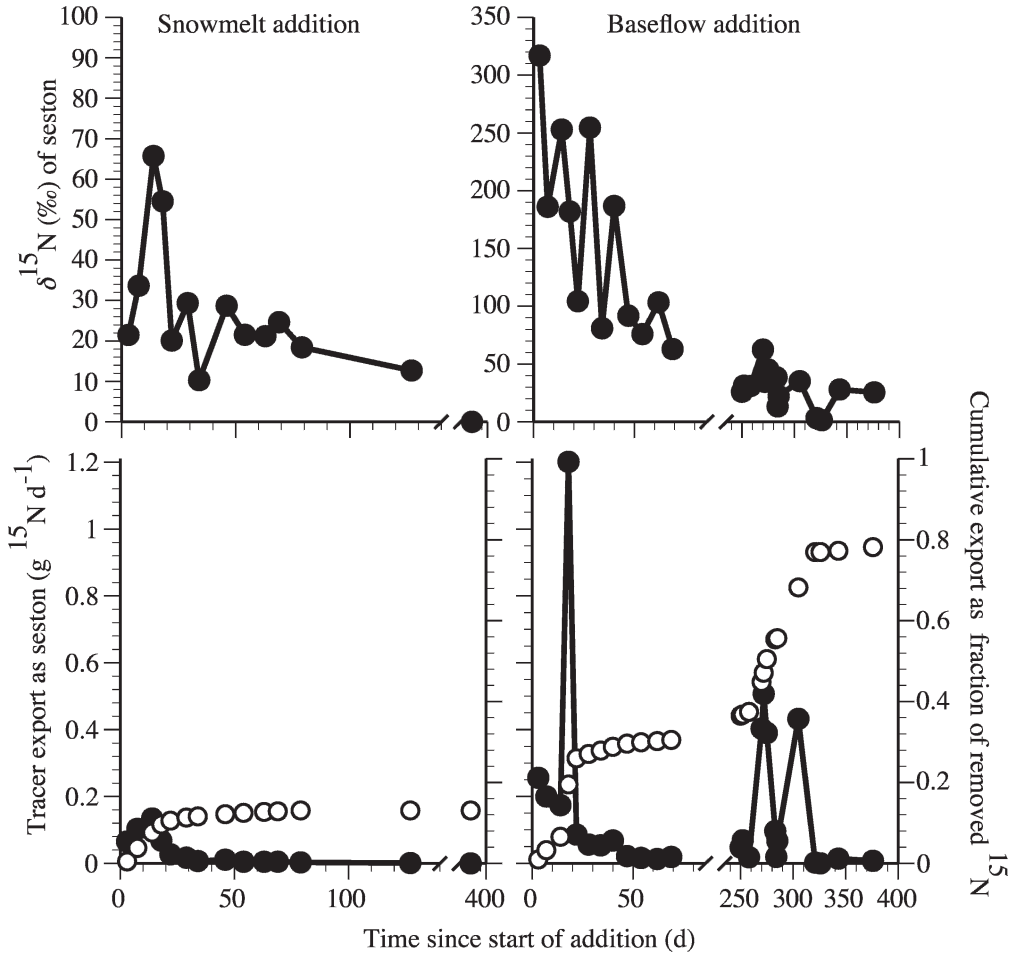

Fig. 9. Much of the stored ^{15}N was exported from the stream as seston. The top two panels show the δ^{15}N of seston through time for the snowmelt and baseflow additions. The bottom panels show both instantaneous export (solid circles, left-axes) and cumulative export (open circles, right-axes). Units for instantaneous export are g ^{15}N d^{-1}, and cumulative export is the fraction of ^{15}N relative to the total amount of ^{15}N removed by the stream.

snowmelt was more likely due to groundwater–surface water exchange, and not because of biological demand on the stream bed. This hyporheic loss of N during snowmelt probably explains why we were able to account for little of the tracer as exported particles. Because of high hydrologic connectivity between the stream and its riparian zone during snowmelt, most of the ^{15}N likely ended up far from the stream. Conversely, during the baseflow addition, it is more likely that the ^{15}N tracer was located in zones closer to the channel where particulate tracer could be more easily

Table 3. Mass balances of nitrogen for the snowmelt and baseflow additions on the final day of the ^{15}N addition (day 14) and the summers following each addition. We added 70 g ^{15}N for the snowmelt addition and 75 g ^{15}N for the baseflow addition. Percent removed is the % of the $^{15}N_{xs}$ that was removed from the water column to the benthos and hyporheic zone during the 2-week addition. Because we found little export as dissolved N after the addition was shut off, we assumed that the cumulative amount of dissolved loss was that which was exported by day 15.

	Snowmelt				Baseflow			
	Day 14 (2002)		Following summer (2003)		Day 14 (2003)		Following summer (2004)	
^{15}N pool	g$^{15}N_{xs}$	% of removed	g$^{15}N_{xs}$	% of removed	g$^{15}N_{xs}$	% of removed	g$^{15}N_{xs}$	% of removed
Amount of tracer exported	45.5	—	—	—	31.5	—	—	—
Amount removed by stream	24.5	100	—	—	43.5	100	—	—
Inventoried in stream channel	1.1	5	0.03	0.1	14.9	34	5.0	12
Exported as particles	1.5	6	2.7	11	2.6	6	31.0	71
Exported as dissolved	0.5	2	0.5	2	0.6	1	0.6	1
Missing	21.4	87	21.3	87	25.4	58	6.9	16

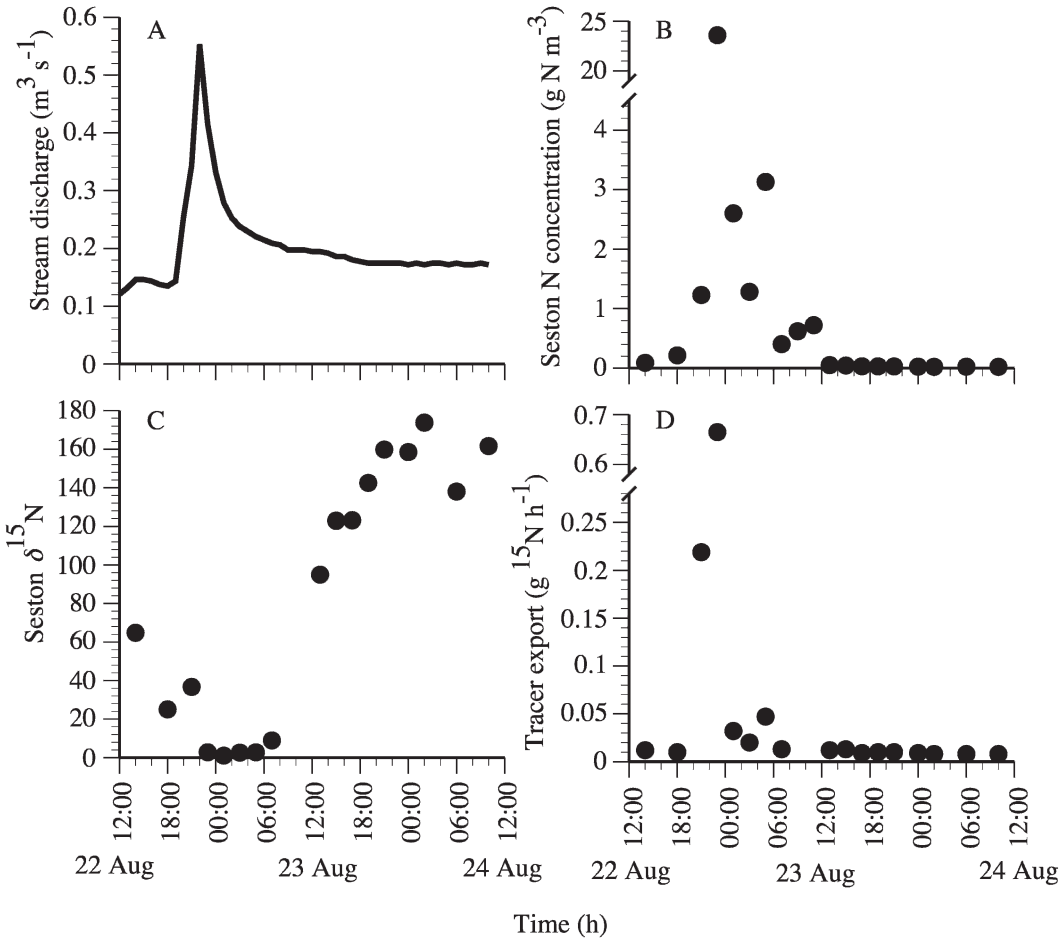

Fig. 10. A storm 3 d after the baseflow addition ended removed a small fraction of the tracer ^{15}N standing stock. (A) shows the storm hydrograph. (B) shows that particulate N concentration (g N m^{-3}) increased during the storm. (C) shows that the $\delta^{15}N$ of seston decreased during the storm, and (D) shows export of $^{15}N_{xs}$ as seston (g^{15}N h^{-1}).

mobilized during high flows (Fig. 9). Our Br^- data (Fig. 4) support this notion in that near-channel exchange was stronger during baseflow compared to snowmelt.

We have five lines of evidence that suggest that the hyporheic zone was where much of the ^{15}N was stored: (1) Based on water-table elevation, there was a spatially consistent net flow of water from the stream to the hyporheic zone. This flow would have carried N from the stream to the riparian zone, where it could be immobilized by soil microbes or vegetation. (2) We measured high standing stocks of particle ^{15}N in the near-stream parafluvial zone. This region contained about 25% of the measurable benthic $^{15}N_{xs}$ standing stock, but it probably only constituted a small fraction of the hyporheic zone. Given that the hyporheic zone was much larger than what we sampled, it likely contained much more immobilized ^{15}N. (3) We measured ^{15}N tracer in streamside willows, showing unequivocally that some tracer was transported away from the stream channel into riparian vegetation. (4) Groundwater wells lateral to the stream channel contained 5–40% stream water as measured by Br^- concentration, and the magnitude of stream water in the subsurface increased with distance from the release point during both tracer tests. (5) Hydrometric analysis from four discharge gauging stations maintained during 2004 suggested that the lower portion of Spring Creek study reach was losing water during both snowmelt peak-flow and summer storms. Comparison of mean annual flows, however, suggested that the stream was either slightly gaining or neutral (Arp et al. 2006). Thus the loss and exchange of water with the riparian floodplain in our study reach varied with flow conditions. Additionally, transient storage was high in the reach with transient storage area/channel cross section area (A_s/A = 0.64) suggesting high exchange of water with backwater or hyporheic zones (Arp and Baker 2007).

Low denitrification rates were also not surprising given low ambient NO_3^- and dissolved organic carbon concentrations (Arp and Baker 2007). Further, the denitrification potential rates we measured in Spring Creek were an order of magnitude lower than ambient rates in parafluvial groundwater of a eutrophic river (Baker and Vervier 2004). While denitrification certainly accounts for some amount of N loss in our study (Mulholland et al. 2008) ambient rates were likely too low to detect significant change in the isotopic composition of the dissolved gas pool.

Long residence time in benthic and hyporheic pools—Residence time of N stored on the stream bed and in the near-hyporheic zone was much longer than those reported from other N isotope tracer studies in streams. We compared residence time of FBON, epilithon N, and wood N with six studies from the Lotic Intersite Nitrogen eXperiment (LINX 1) and an isotope addition to an estuary (Dodds et al. 2000; Tank et al. 2000; Hamilton et al. 2001; Mulholland et al. 2001; Merriam et al. 2002; Tobias et al. 2003; Ashkenas et al. 2004). Residence times for those studies averaged 26 d (range = 7–53 d) for FBON, 20 d (range = 1.5–47 d) for epilithon, and 33 d (range = 19–47 d) for wood. Residence times of any pool in Spring Creek were much longer than the longest residence times from the LINX 1 study. The shortest residence time we were able to measure was 117 d for both epilithon and wood biofilm during the baseflow addition. FBON residence times were $>$ 200 d, and hyporheic N exceeded 1 yr because hyporheic standing stocks of ^{15}N were not depleted 1 yr after the baseflow addition. These residence times were longer than those for an estuary where 60% of stored N was in sediment samples and observed $\geq$ 2 months following the isotope addition (Tobias et al. 2003). The cold climate in Spring Creek may have contributed to longer storage of N, but even if this stream were as biologically active all year as during the 6-month growing season, it would still have much longer residence times for N than the LINX streams because residence times were twice as long as the values from other streams.

Year-scale retention of isotope has been noted in only a few studies. Peterson et al. (1997) found labeled insects and epilithon in the Kuparuk River one summer later. The Kuparuk freezes solid from October to May, thus shutting down mineralization and fluvial export. Insects have 2–3-yr life cycles, which will promote interannual storage of ^{15}N in their tissue (Peterson et al. 1997). In Mack Creek, Oregon, mosses and epilithon each contained 5% of their peak label 1 yr following ^{15}N addition (Ashkenas et al. 2004).

Our data suggest that uptake in the hyporheic zone is an important mechanism by which N can be stored for long periods as shown for the hyporheic zone in Mack Creek where riparian plants were enriched following ^{15}N addition (Ashkenas et al. 2004). Streams similar to Spring Creek, at mountain fronts with valley and slope transitions, gain and lose water over long (100–1000 s of m) spatial scales (Covino and McGlynn 2007). A similar process likely operates here to affect N retention in the subsurface, because of the hydrogeomorphic transition of a stream approaching a lake and its delta (Arp et al. 2006, 2007).

If in fact the hyporheic zone was a primary storage zone for particulate N as the hydrologic data and labeled willows suggest, then it posits a mechanism by which residence time can be so long in some of the shallow benthic pools. Either mineralization followed by transport and reuptake of N by the benthos, or movement of particle N from deeper to shallow pools may have allowed benthic pools to remain enriched for a long time. We could find no direct evidence in the literature to support either mechanism, but Holmes et al. (1994) suggested that stream-derived particles were mineralized in the hyporheic zone and contributed to high nitrification rates and supported algal growth in areas where hyporheic water reentered the stream (Valett et al. 1994; Henry and Fisher 2003).

Floods and seston export—Although most measurements of nutrient cycling occur during baseflow conditions, it is important to recognize that in snowmelt-dominated watersheds, little nutrient transport occurs during baseflow, but rather during high flows (Meyer and Likens 1979; Creed et al. 1996; Baron and Campbell 1997). The form of this N can be dissolved (as NO_3^-; Baron and Campbell 1997) or particulate (Wurtsbaugh et al. 2005).

In Spring Creek, seston dominated ^{15}N export. Other studies have shown that mineralized N (as DON or NH_4^+) can be important losses (Hamilton et al. 2001; Merriam et al. 2002; Tobias et al. 2003). The amount of tracer exported in dissolved pools was low in Spring Creek. In addition, the mass balance for ^{15}N in the baseflow addition shows that most of the ^{15}N-NO_3^- removed by the stream was subsequently accounted for as seston (Table 3).

Despite that nearly all ^{15}N export was as seston, summer floods scoured little of the labeled N from the stream. The summer spate that occurred 3 d following the baseflow addition scoured 16% of the $^{15}N_{xs}$ found on the stream bed and constituted 8% of the total seston $^{15}N_{xs}$ export. This resistance to particle export occurred despite gravelly bed sediments that are highly mobile during floods (Myers et al. 2007), which we assume would have facilitated organic matter loss during storms. Other stream tracer studies that have had serendipitous floods showed similar resistance to benthic N export. A spate during a ^{15}N addition to a Puerto Rican rainforest stream increased discharge 20-fold, but only removed 37% of the N tracer in stream detrital pools (Merriam et al. 2002). A storm increased discharge 10-fold in Ball Creek, North Carolina, U.S.A. but did not reduce $\delta^{15}N$ in benthic pools (Tank et al. 2000). At first glance, these results suggest a paradox. Tracer studies show that benthic N is resistant to flooding (Tank et al. 2000; Merriam et al. 2002; our data), yet budget studies show that most export occurs during storms (Webster et al. 1990). Using our mass balance approach, we suggest these two findings are congruent. Because of the strong decline in $\delta^{15}N$ during the storm, much of what was exported during the flood was not from the labeled benthos in the study reach, but from upstream, the unwetted channel, or from upland sources. With moderate storms, a fraction of stream benthic $^{15}N_{xs}$ is exported and this amount is larger than what would have been exported over the same time interval during baseflow. However, the bulk of $^{15}N_{xs}$ stored was resistant to export by any one storm. Over long timescales (and several floods) much of the benthic N in Spring Creek will eventually be winnowed out as fine particles.

A longer term view of stream element cycling—In order to understand and predict how streams transport and transform elements such as N, we show that it is necessary to account for these processes over time and over a range of environmental variability (i.e., snowmelt floods, rain storms, and baseflow). Ecologists have a body of theory and data showing mechanisms of N uptake across a range

of streams (Tank et al. 2008; Hall et al. 2009), but we have not incorporated hydrologic variability into this theory, despite knowing for 30 yr that hydrologic variation regulates element export (Meyer and Likens 1979). Additionally, it is necessary to consider the stream as part of a larger parafluvial and riparian ecosystem because elements in transport do not necessary remain in the channel (Fisher et al. 1998). Hyporheic exchange represented an important nitrate sink, especially during snowmelt when benthic biomass and uptake was low. The fate of this missing N is unknown, but we hypothesize that this N is in long-term soil and vegetation pools where residence times of N will be long simply because the pools are so large (Likens and Bormann 1995). In this regard, we consider streams with a large connected floodplain or hyporheic zone to be analogous to a small watershed (Bormann and Likens 1967; Likens and Bormann 1995). Over long timescales (millennia) streams with floodplains are probably in steady state with respect to N cycling (Brookshire et al. 2009), but over shorter timescales (months–years), they may not be in steady state because the storage pool is so large that slight changes to the size of this pool could dramatically alter export. Incorporating hyporheic storage and hydrologic variation in our model of N cycling, we suggest that removal of N from the water column is decoupled from hydrologic export (i.e., exported N is not from the same time or place as N removed from streamwater). Therefore, Spring Creek is not in steady state with respect to its nutrient budget at months–years timescales, similar to phosphorus in Bear Brook (Meyer and Likens 1979).

There is much current interest in the role of streams and rivers in removing N (Alexander et al. 2000; Seitzinger et al. 2002). Denitrification is thought to be the primary process in which streams and rivers remove N from transport (Seitzinger et al. 2006; Mulholland et al. 2008), though, on average, 84% of NO_3^- removal is via assimilation (Hall et al. 2009; Mulholland et al. 2009). We show that storage in the stream bed can retard N export, and may be an important fate, especially if streams have intact hyporheic zones and floodplains. Additionally, assimilatory uptake may precede denitrification in riparian zones. Contrary to our hypothesis, we show that Spring Creek was not solely a conduit for nutrients at high flows, but had as high an uptake velocity for N during the snowmelt flood as during summer baseflow. Given that most dissolved nutrient transport occurs at high flows (Baron and Campbell 1997), streams with connected hyporheic zones or floodplains may be able to absorb some of this N at a time when biotic uptake is low. Variable flows may promote dissolved nutrient transformation and storage in streams with connected hyporheic zones and floodplains.

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